Figure 1: Various synthetic pathways for the biosynthesis of DHA (docosahexaenoic acid)

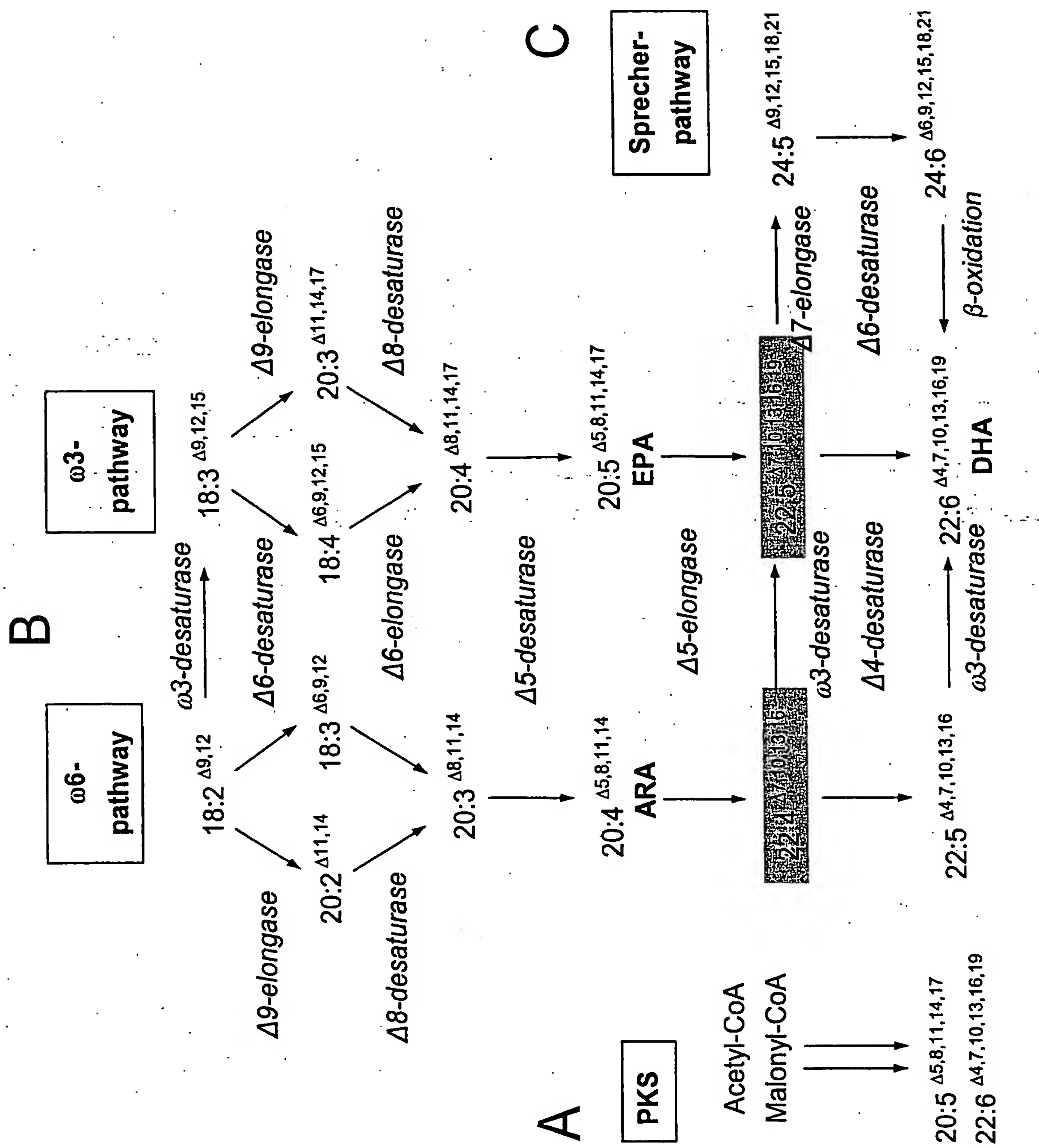


Figure 2: Substrate specificity of the Δ5-elongase (SEQ ID NO: 53) with regard to different fatty acids

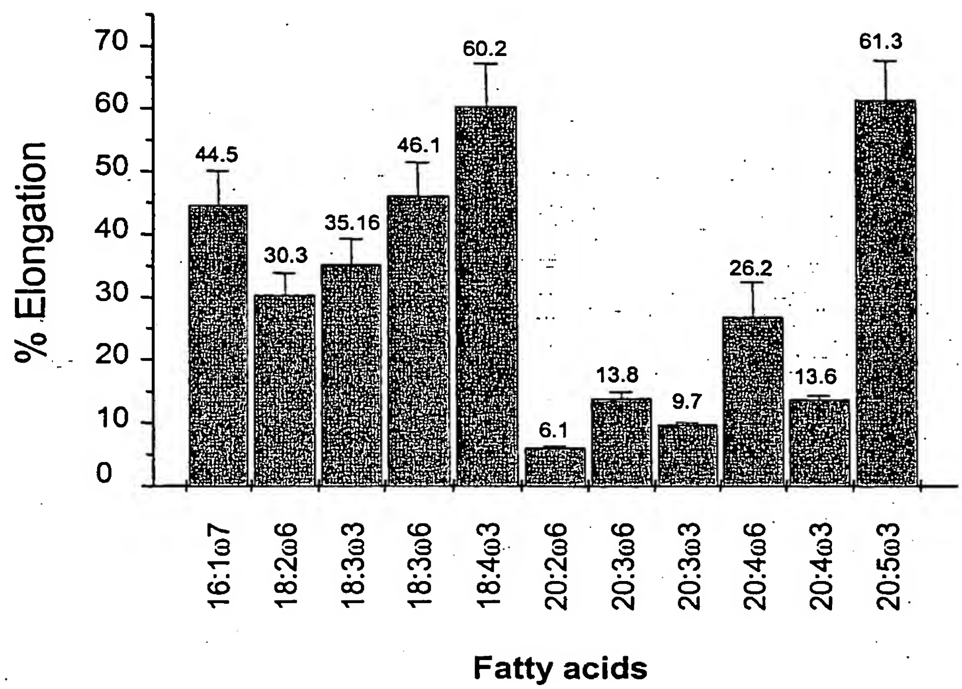


Figure 3: Reconstitution of DHA biosynthesis in yeast starting from 20:5ω3.

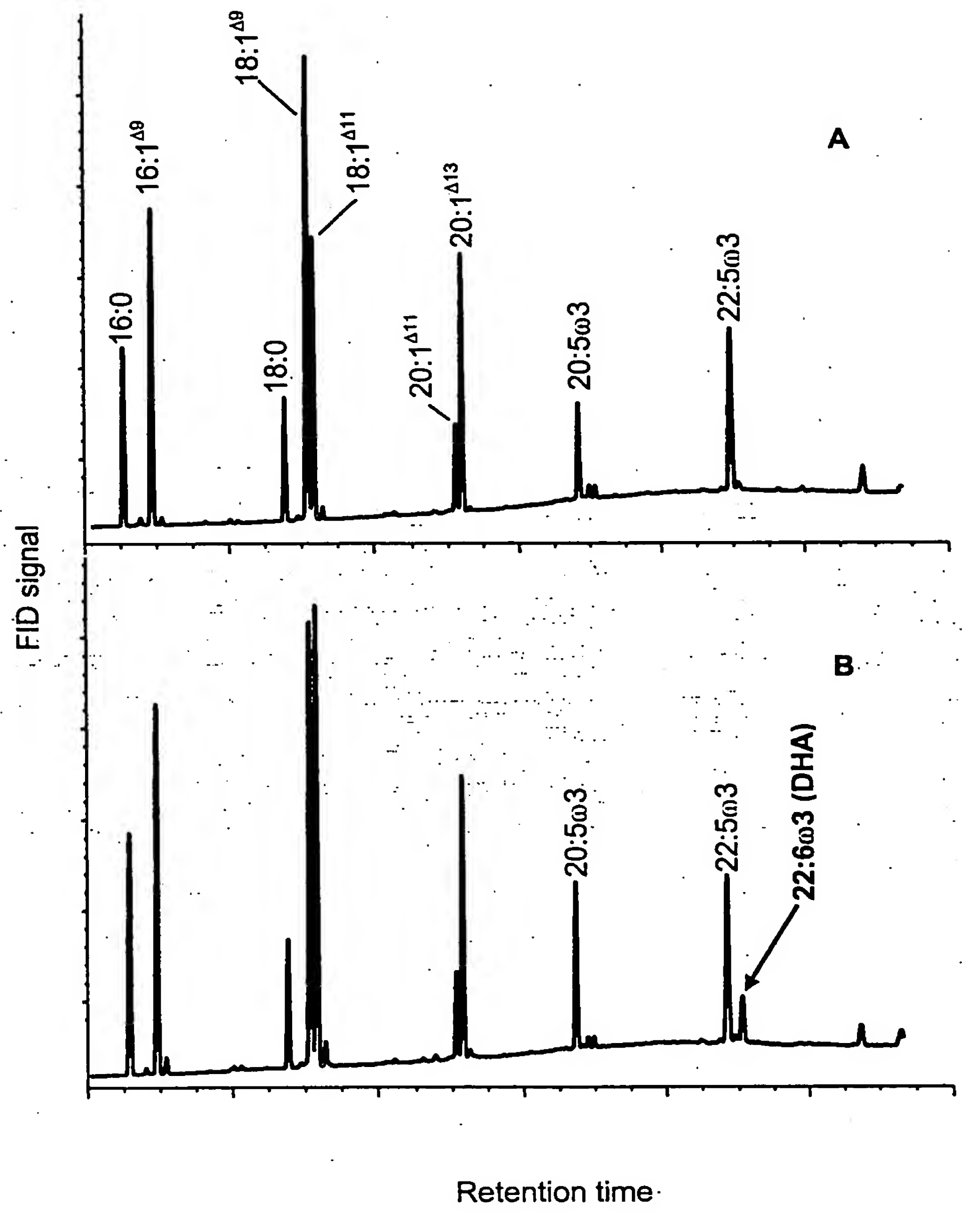


Figure 4: Reconstitution of DHA biosynthesis in yeast starting from 18:4ω3.

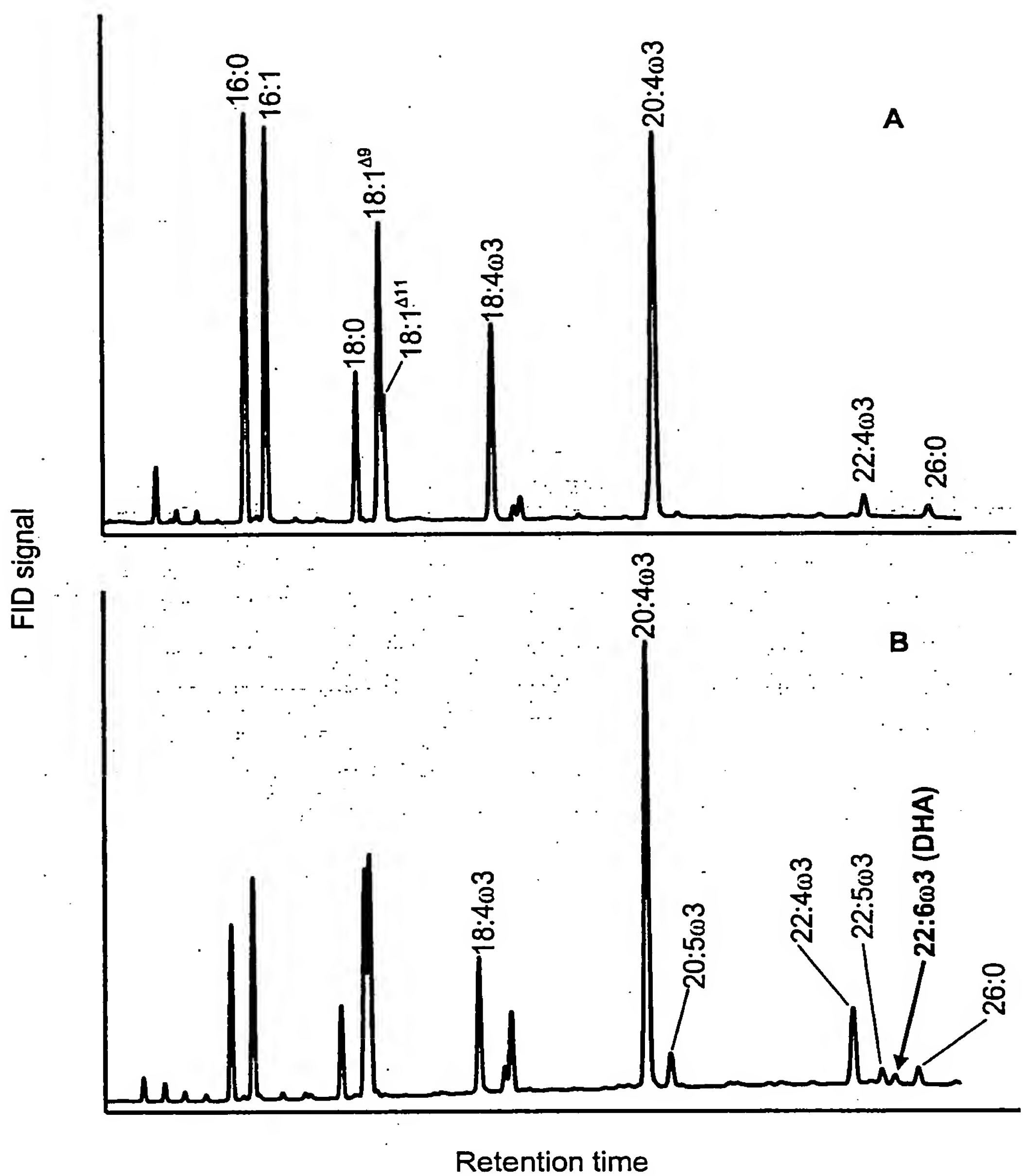


Figure 5: Fatty acid composition (in mol%) of transgenic yeasts which had been transformed with the vectors pYes3-OmELO3/pYes2-EgD4 or pYes3-OmELO3/pYes2-EgD4+pESCLeu-PtD5. The yeast cells were cultured in minimal medium without tryptophan and uracil/ and leucin in the presence of 250 μM $20:5^{\Delta5,8,11,14,17}$ and $18:4^{\Delta6,9,12,15}$, respectively. The fatty acid methyl esters were obtained from cell sediments by acid methanolysis and analyzed via GLC. Each value represents the mean (n=4) ± standard deviation.

	pYes3-OmELO/pYes2-EgD4	pYes3-OmELO/pYes2-EgD4 EgD4 + pESCLeu-PtD5
Fatty acids	Feeding of $20:5^{\Delta5,8,11,14,17}$	Feeding of $18:4^{\Delta6,9,12,15}$
16:0	9.35 ± 1.61	7.35 ± 1.37
$16:1^{\Delta9}$	14.70 ± 2.72	10.02 ± 1.81
18:0	5.11 ± 1.09	4.27 ± 1.21
$18:1^{\Delta9}$	19.49 ± 3.01	10.81 ± 1.95
$18:1^{\Delta11}$	18.93 ± 2.71	11.61 ± 1.48
$18:4^{\Delta6,9,12,15}$	-	7.79 ± 1.29
$20:1^{\Delta11}$	3.24 ± 0.41	1.56 ± 0.23
$20:1^{\Delta13}$	11.13± 2.07	4.40 ± 0.78
$20:4^{\Delta8,11,14,17}$	-	30.05 ± 3.16
$20:5^{\Delta5,8,11,14,17}$	6.91± 1.10	3.72 ± 0.59
$22:4^{\Delta10,13,16,17}$	-	5.71 ± 1.30
$22:5^{\Delta7,10,13,16,19}$	8.77 ± 1.32	1.10 ± 0.27
$22:6^{\Delta4,7,10,13,16,19}$	2.73 ± 0.39	0.58 ± 0.10

Figure 6: Feeding experiment for determining the functionality and substrate specificity with yeast strains

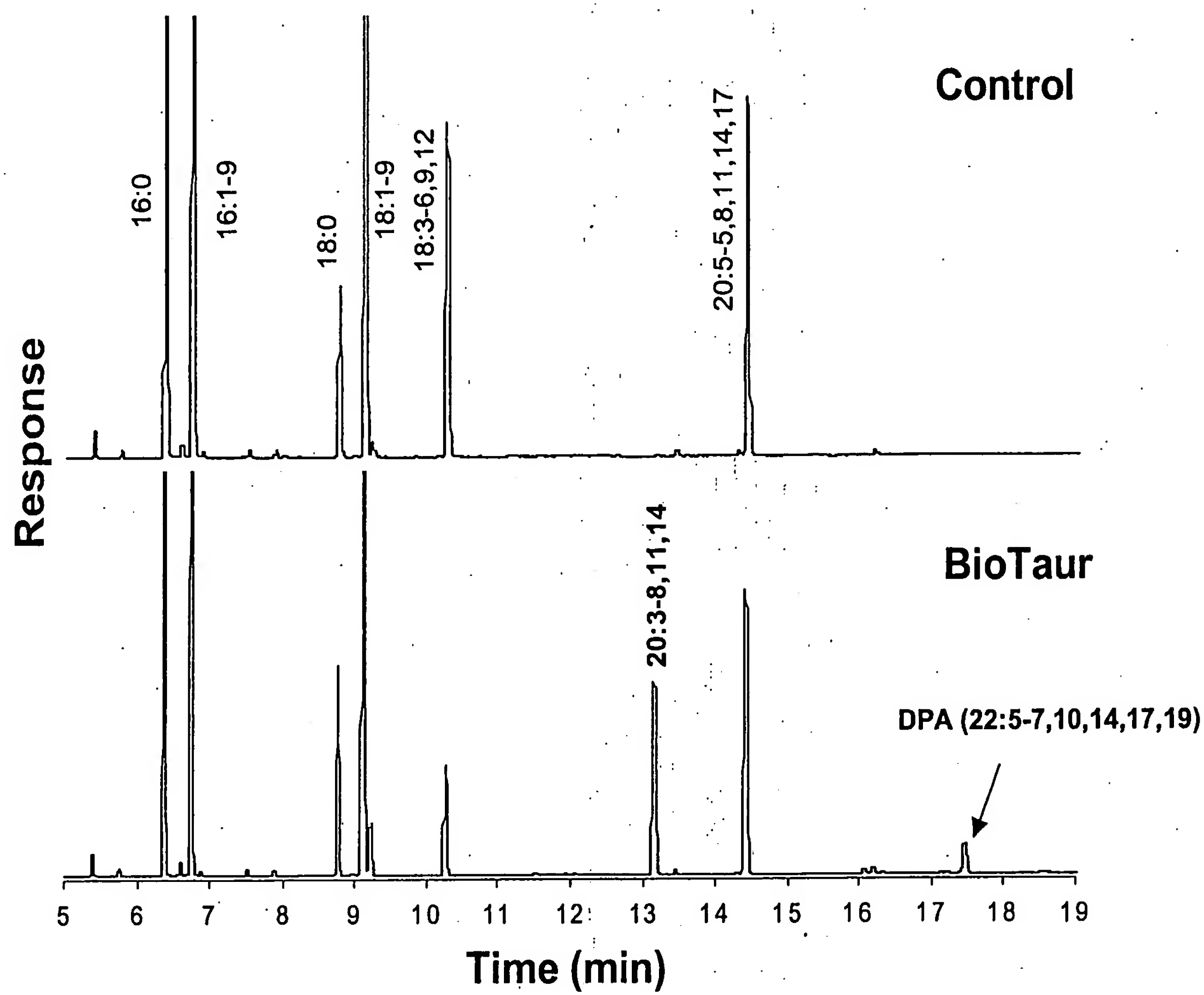
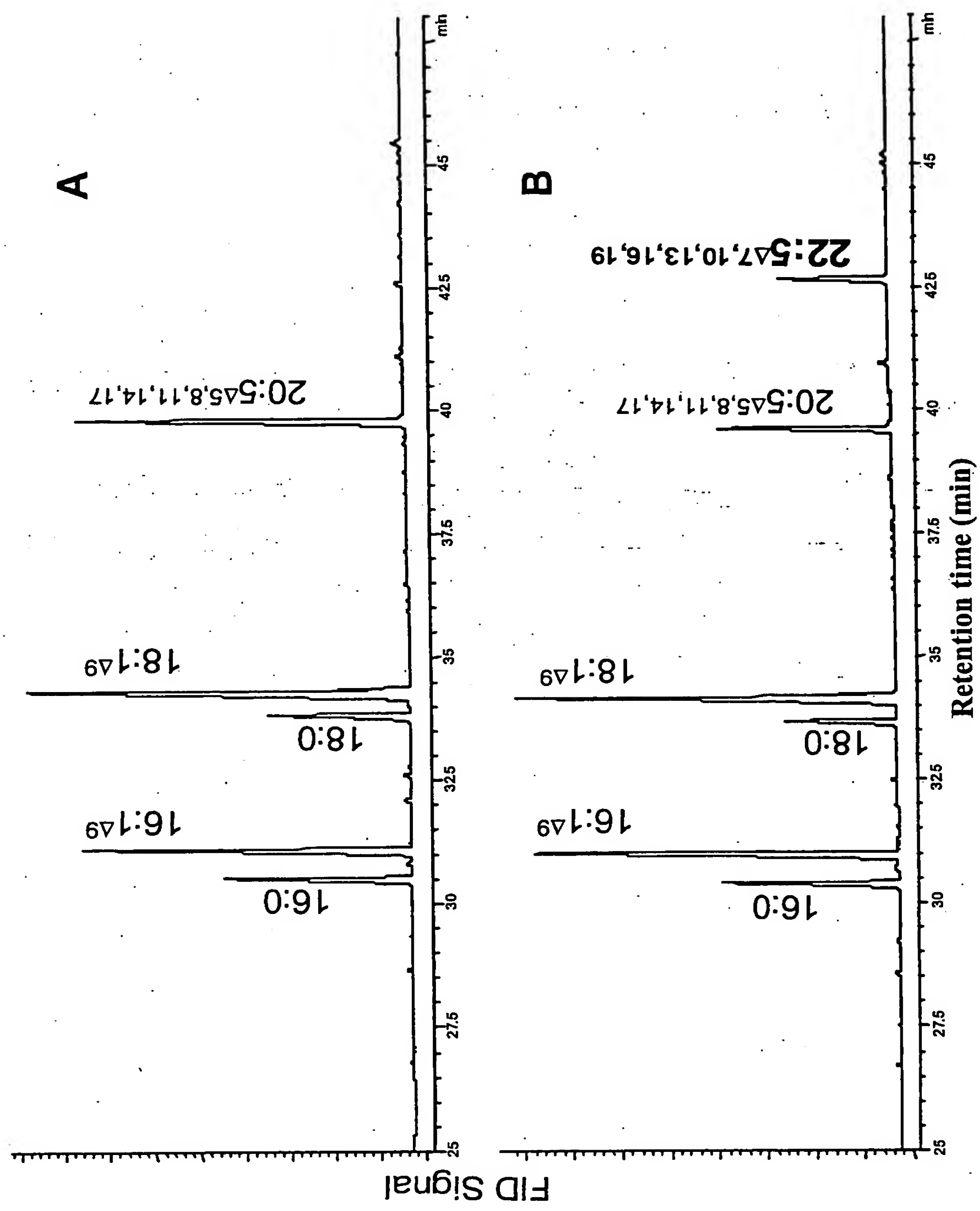


Figure 7: Elongation of eicosapentaenoic acid by OtElo1



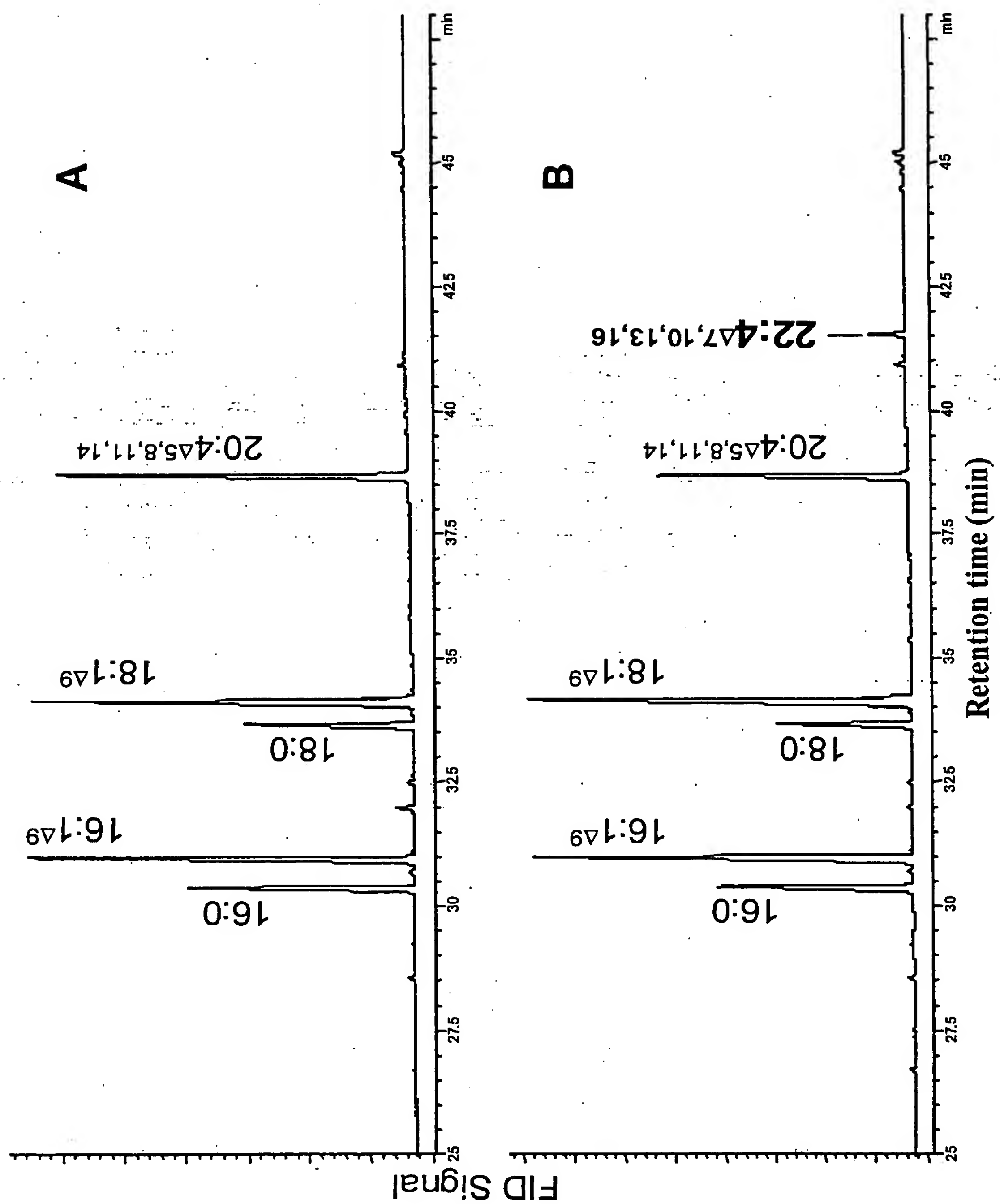


Figure 8: Elongation of arachidonic acid by OtElo1

Figure 9: Expression of TpELO1 in yeast

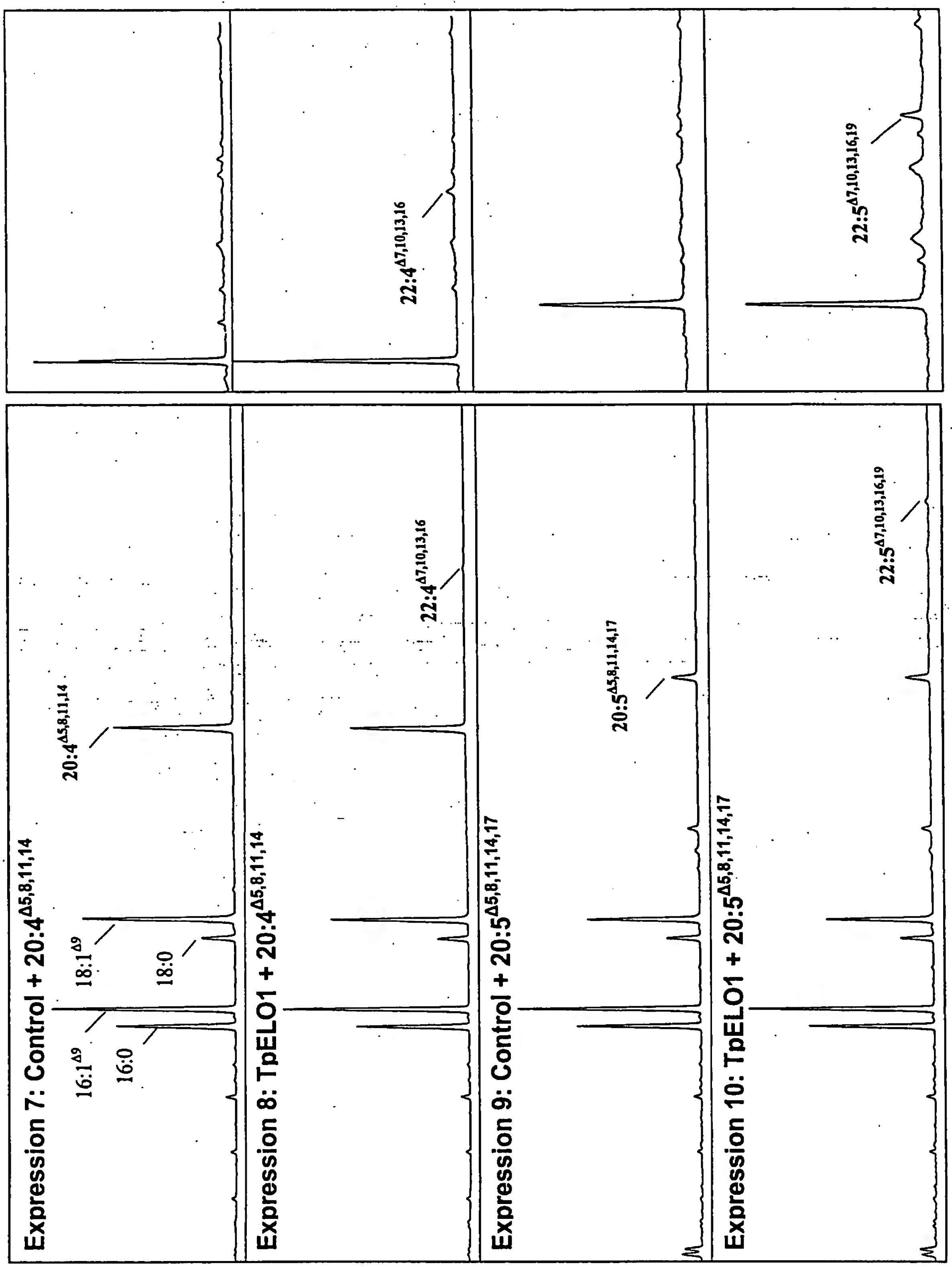


Figure 10: Expression of TpELO3 in yeast

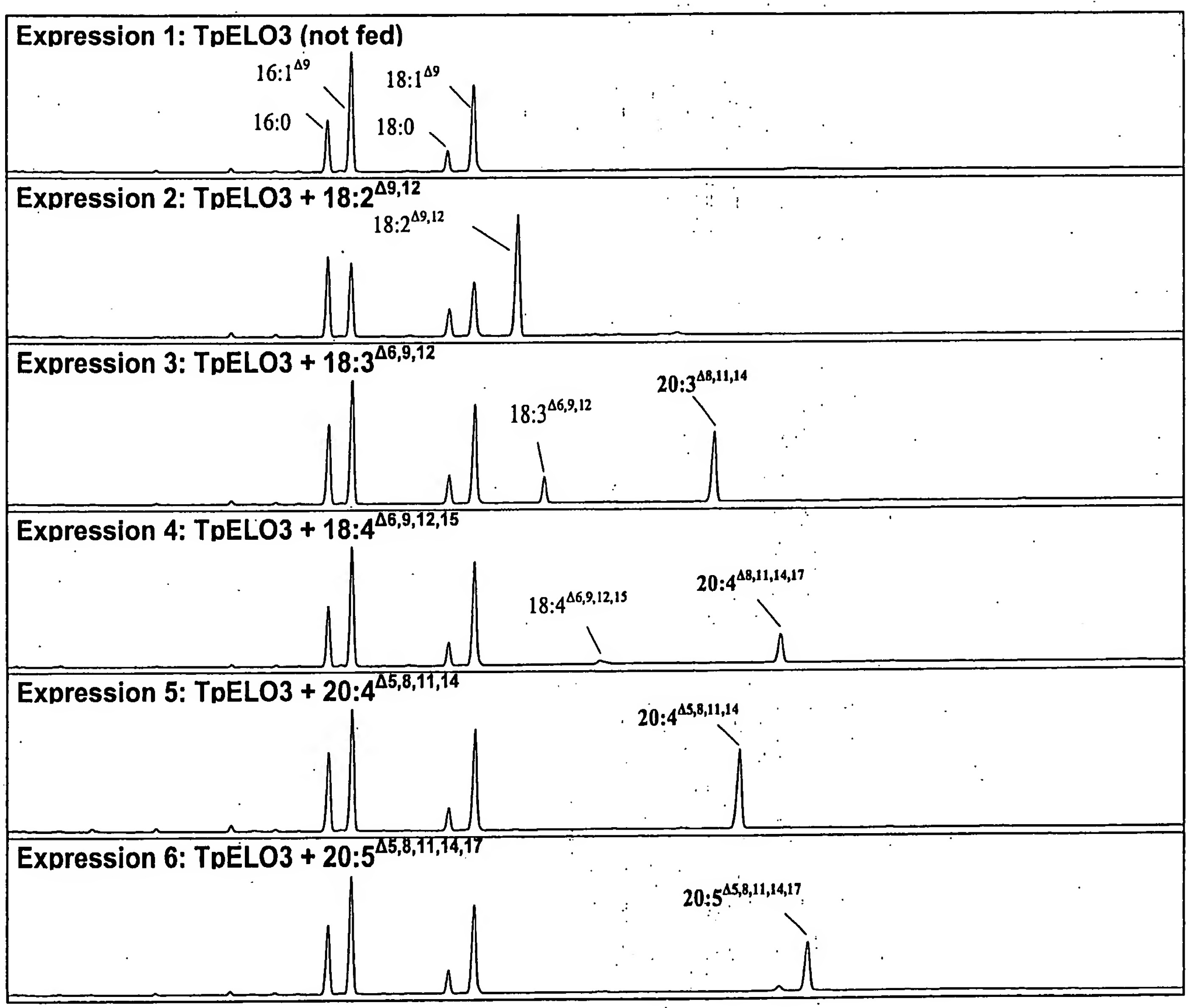


Figure 11: Expression of Thraustochytrium Δ5-elongase TL16/pYES2.1 in yeast

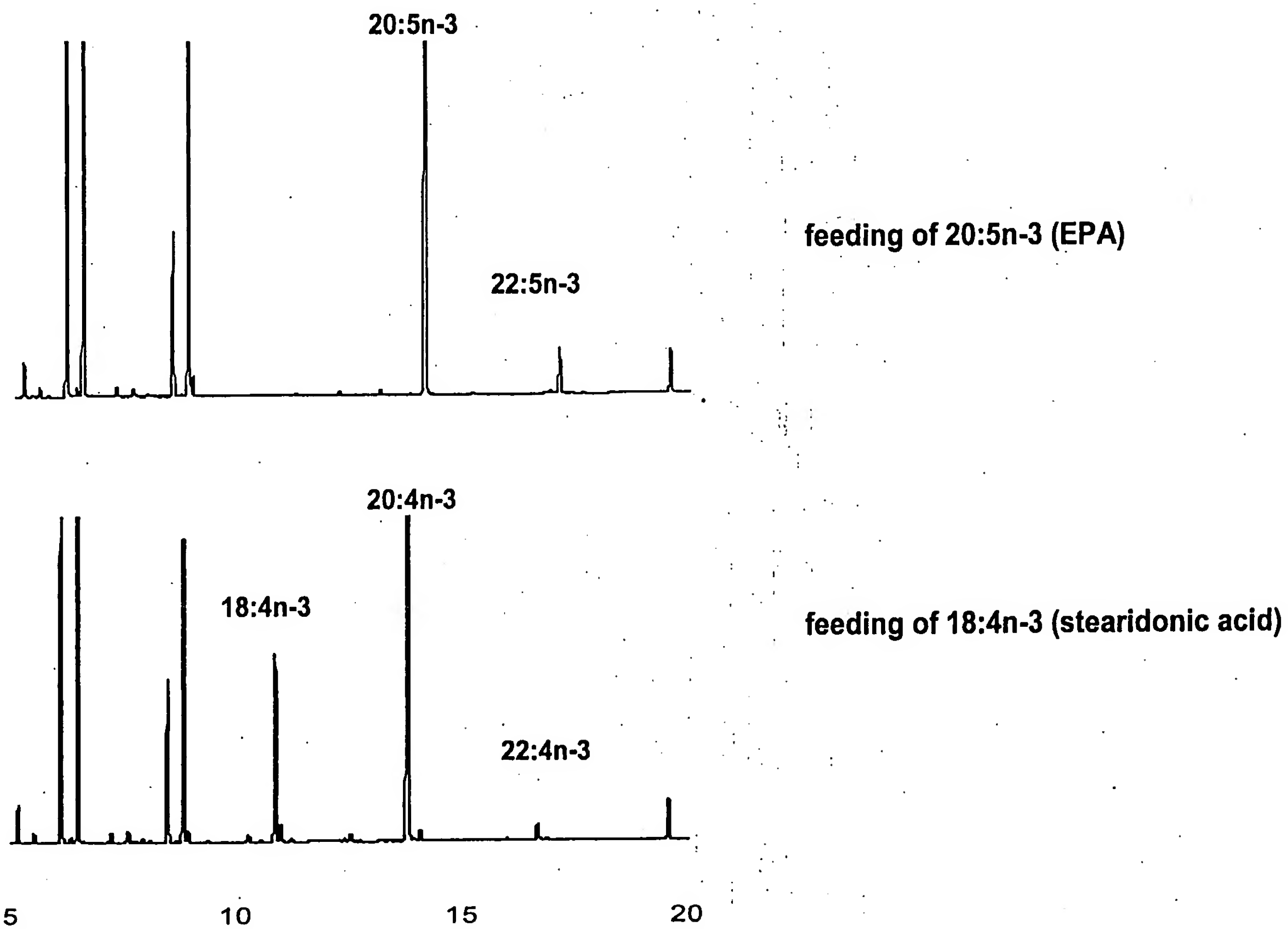


Figure 12: Desaturation of γ-linolenic acid (18:2 ω6-fatty acid) to give α-linolenic acid (18:3 ω3-fatty acid) by Pi-omega3Des.

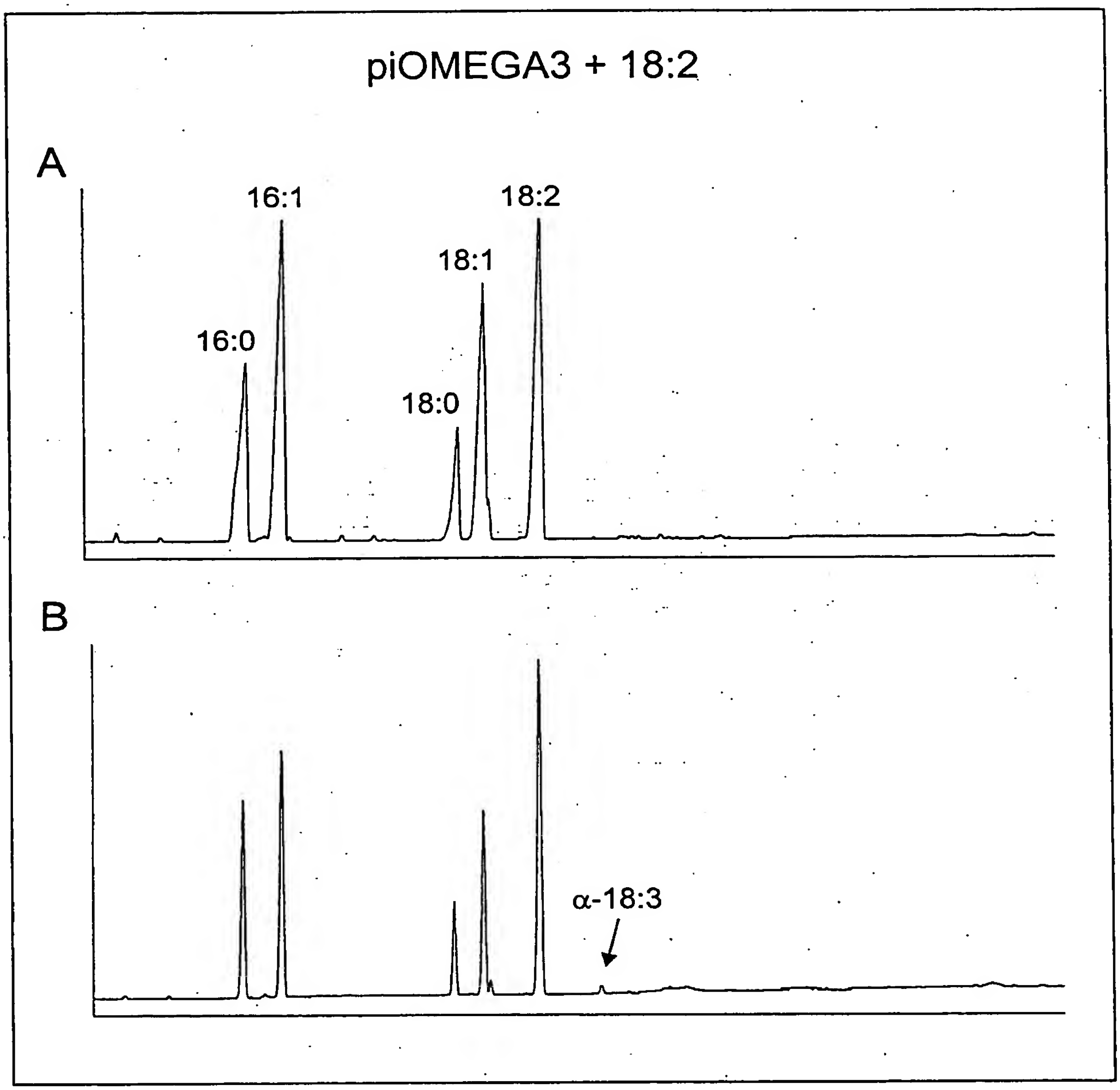


Figure 13: Desaturation of γ-linolenic acid (18:2 ω6-fatty acid) to give stearidonic acid (18:4 ω3-fatty acid) by Pi-omega3Des.

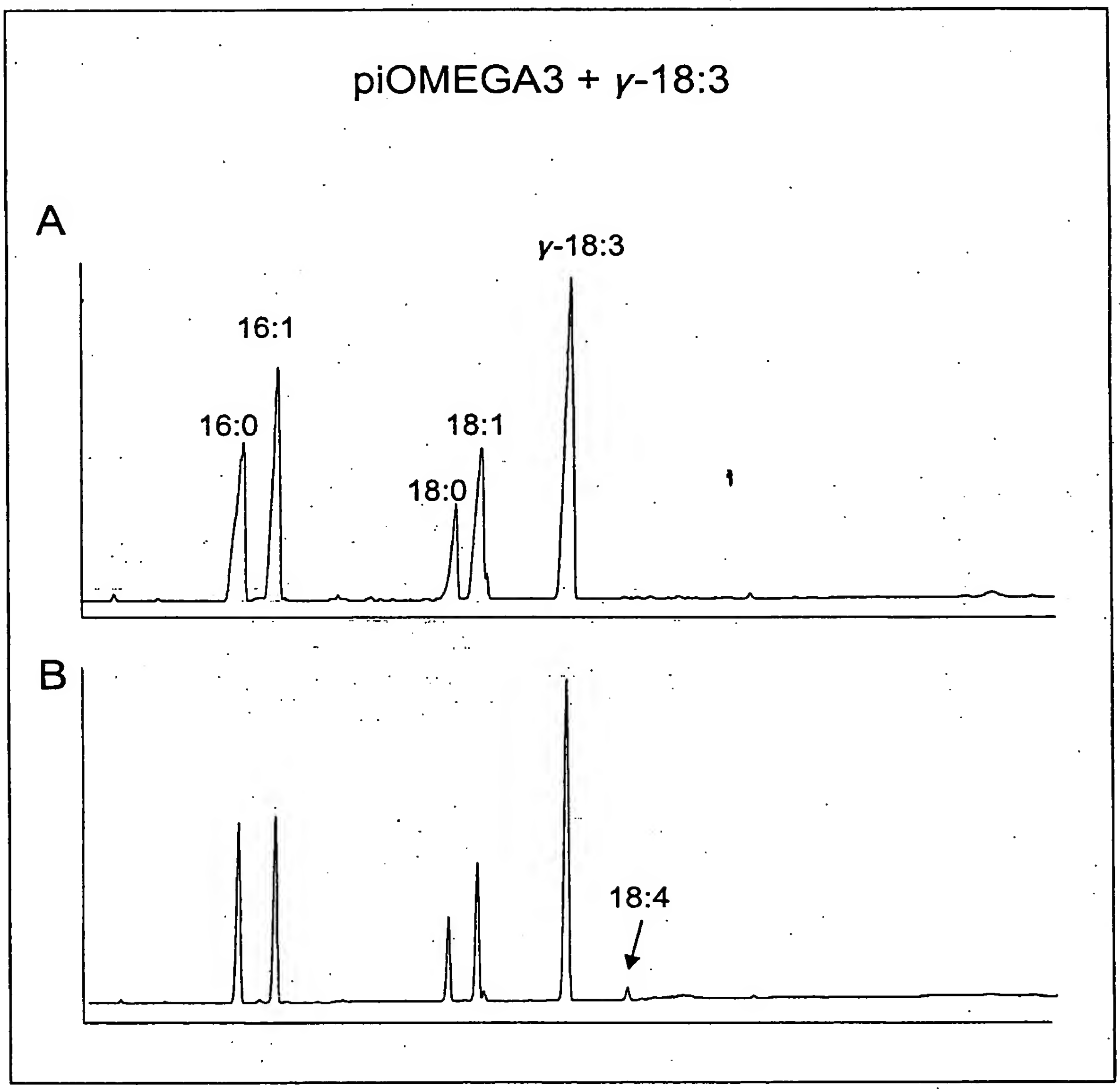


Figure 14: Desaturation of C20:2 ω6-fatty acid to give C20:3 ω3-fatty acid by Pi-omega3Des.

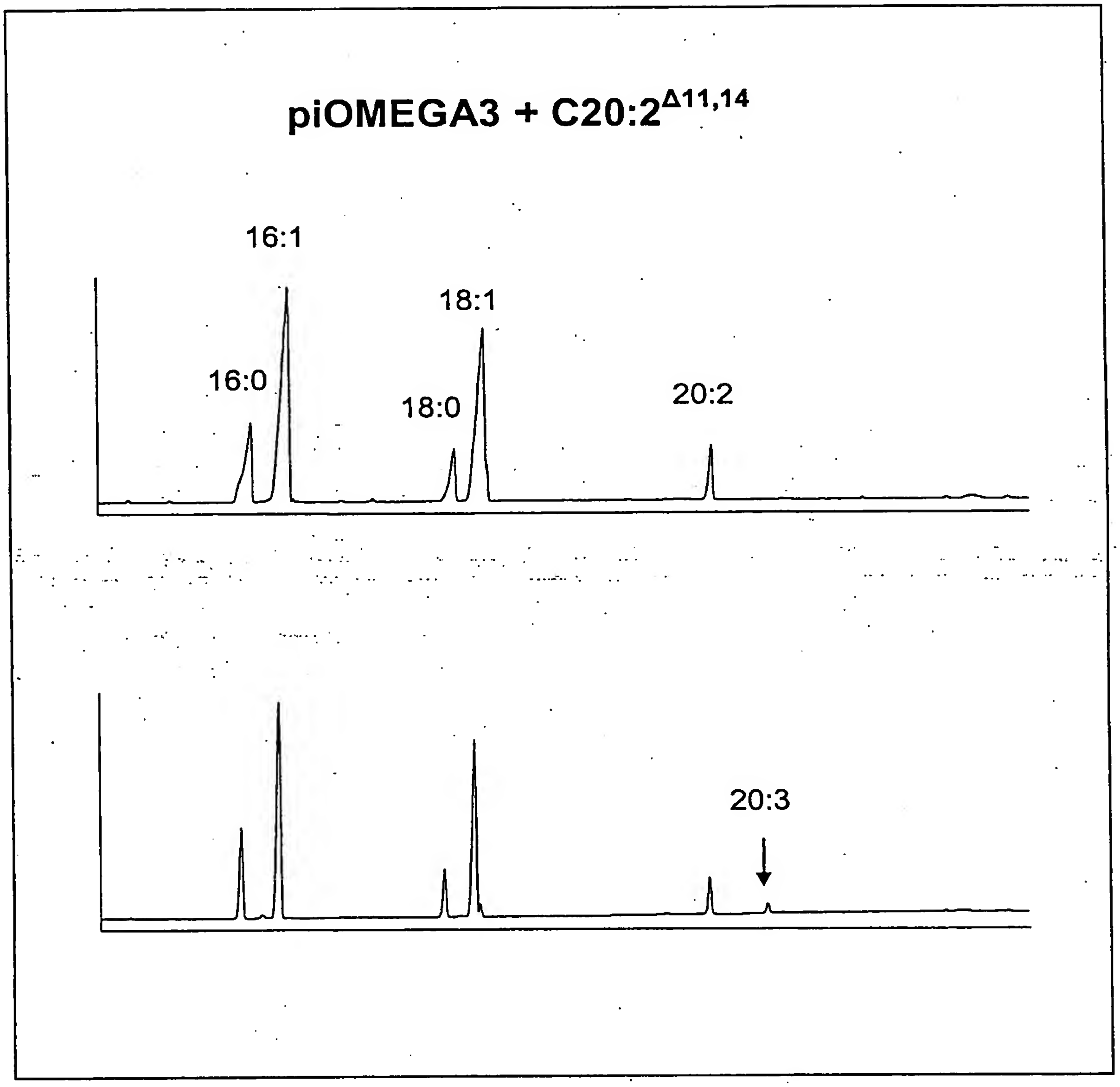


Figure 15: Desaturation of C20:3 ω6-fatty acid to give C20:4 ω3-fatty acid by Pi-omega3Des.

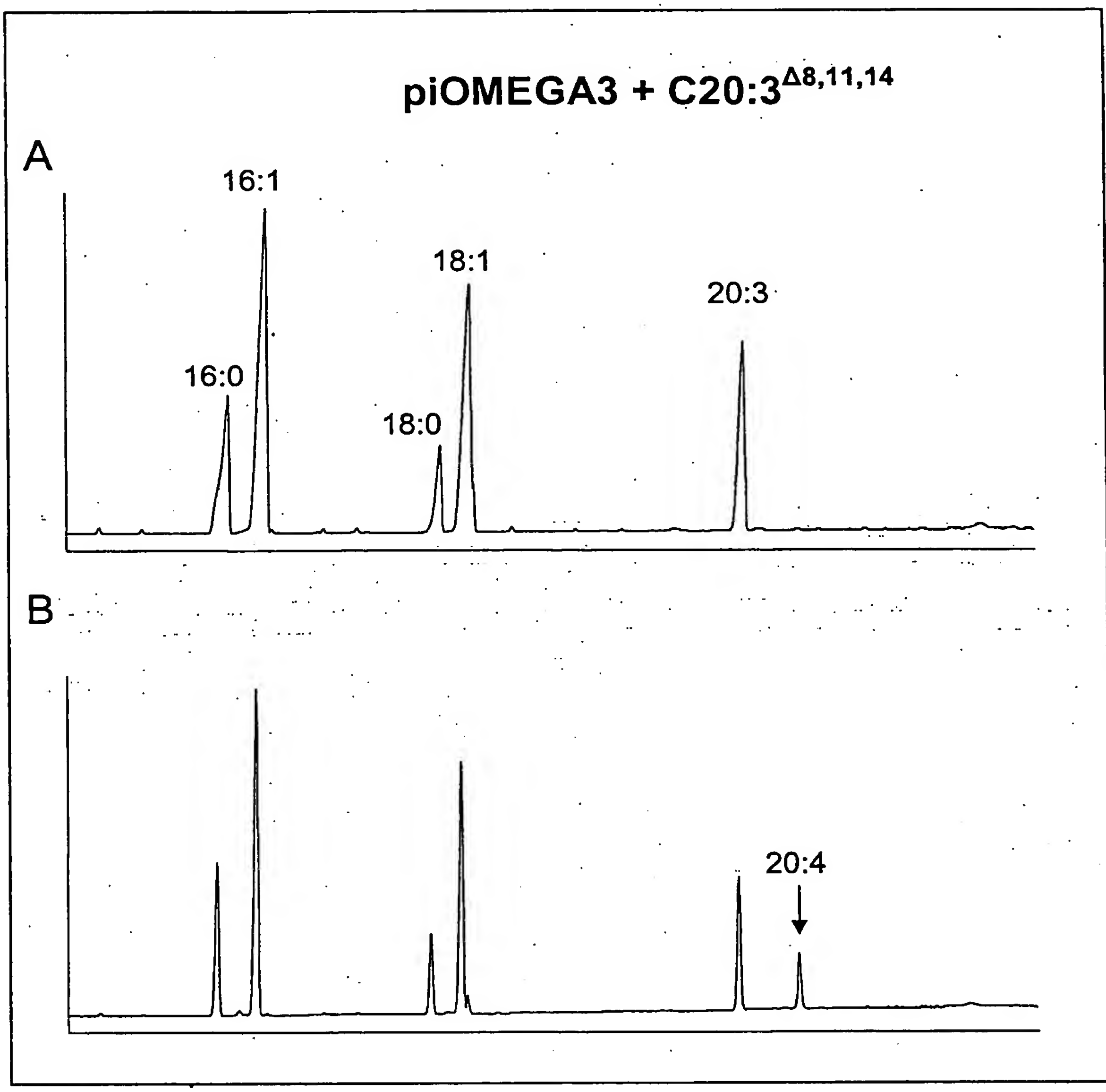


Figure 16: Desaturation of arachidonic acid (C20:4 ω6-fatty acid) to give eicosapentaenoic acid (C20:5 ω3-fatty acid) by Pi-omega3Des.

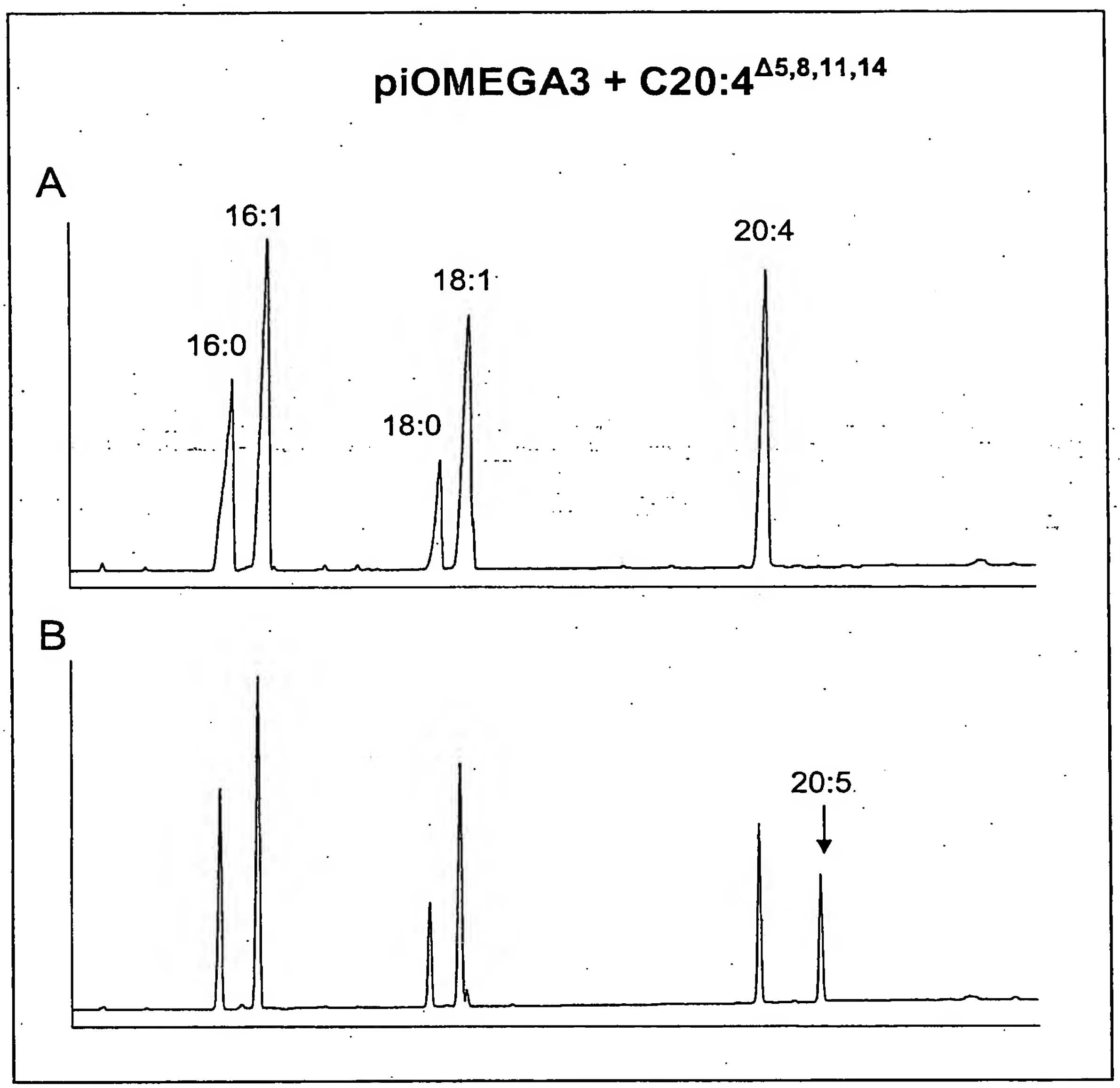


Figure 17: Desaturation of docosatetraenoic acid (C22:4 ω6-fatty acid) to give docosapentaenoic acid (C22:5 ω3-fatty acid) by Pi-omega3Des.

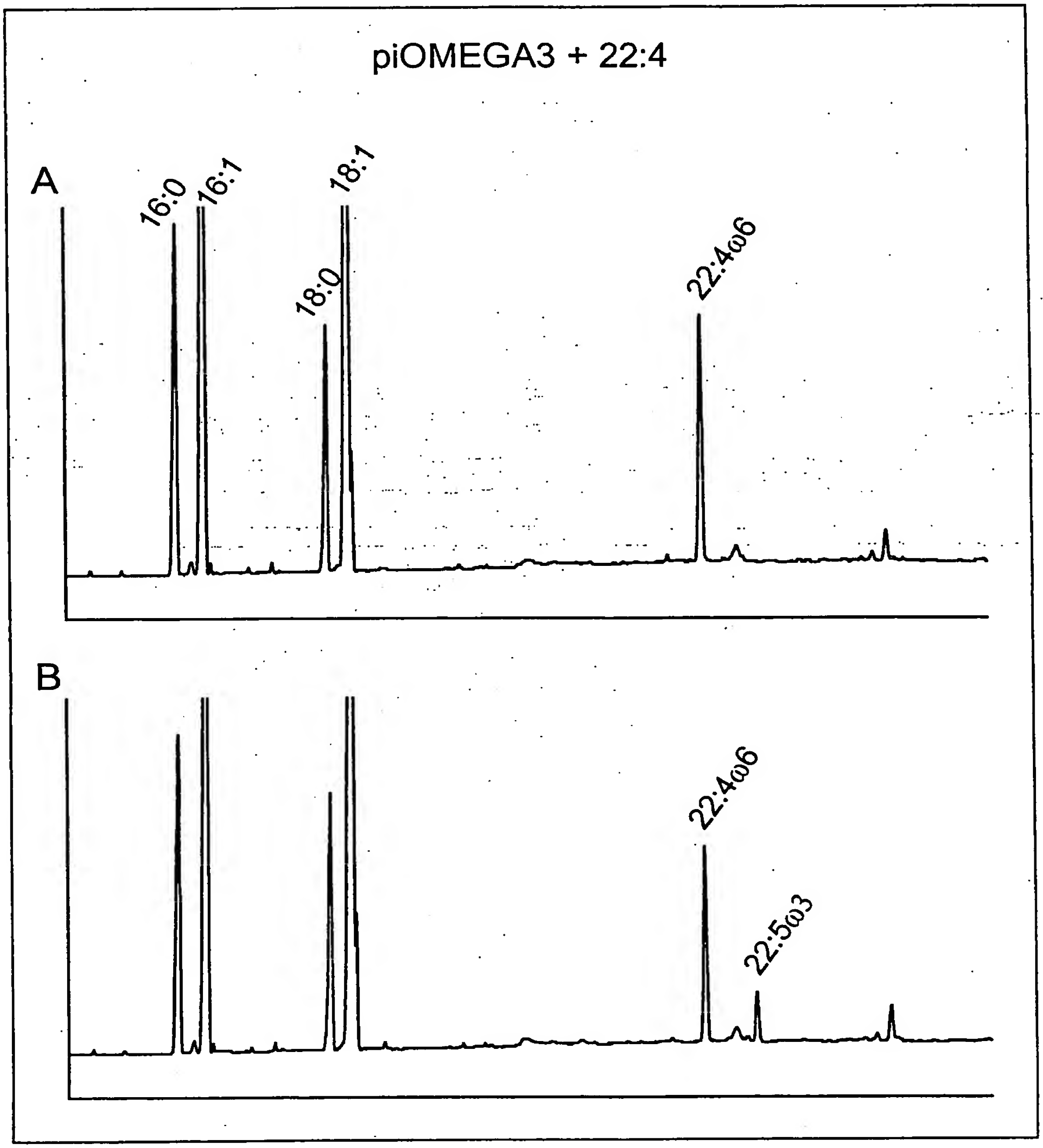
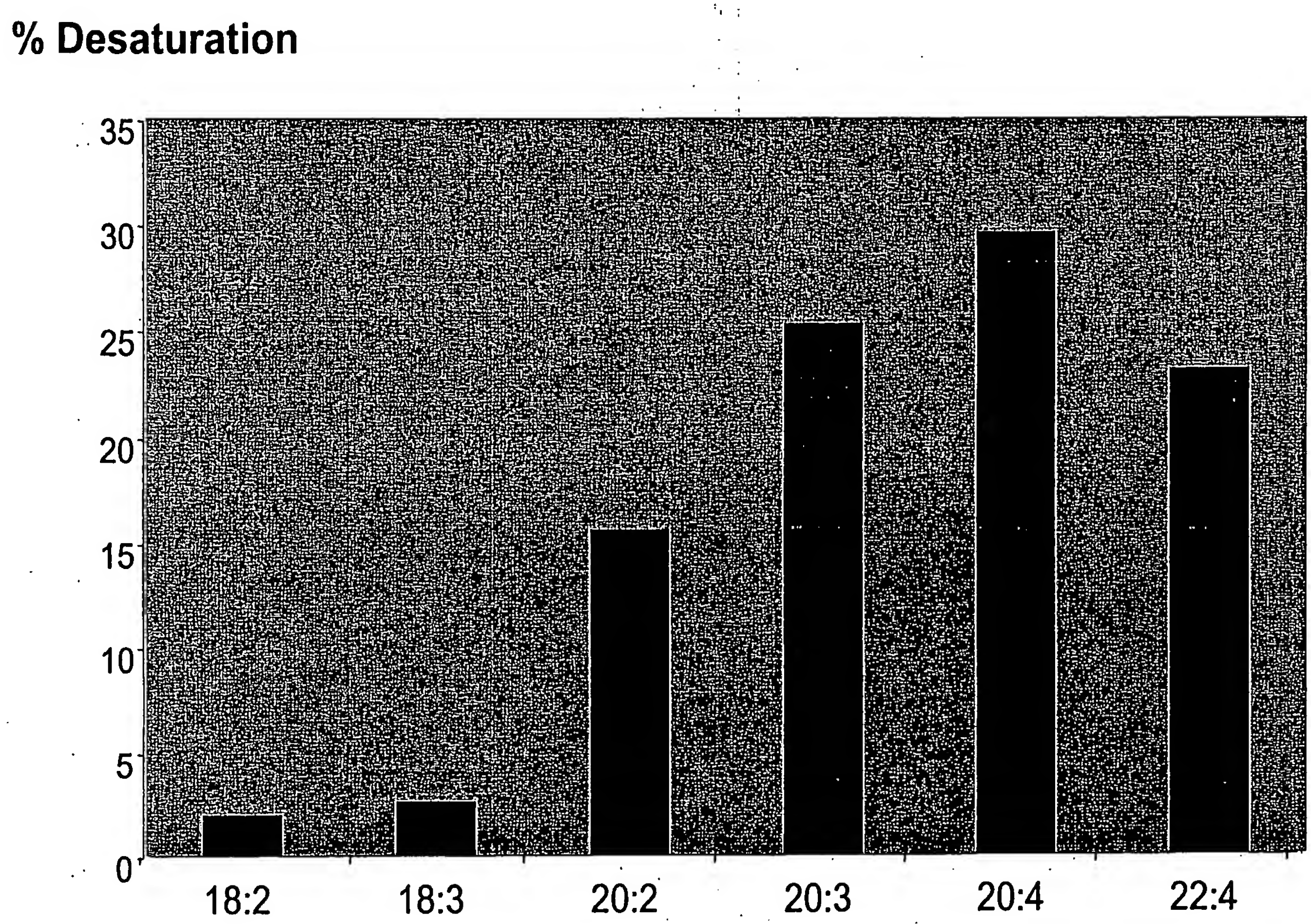


Figure 18: Substrate specificity of Pi-omega3Des with regard to different fatty acids



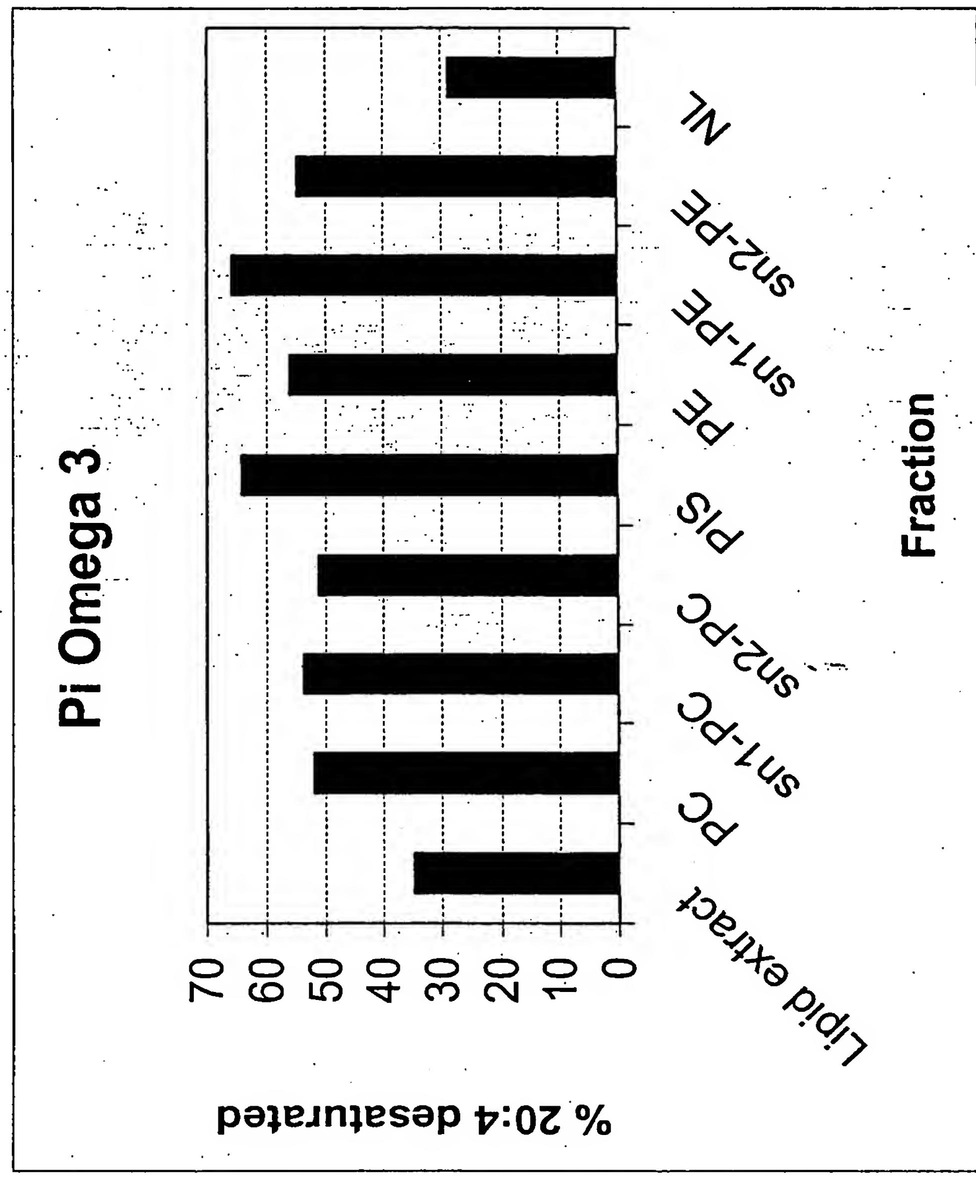


Figure 19: Desaturation of phospholipid-bound arachidonic acid to give EPA by Pi-Omega3Des

Figure 20: Conversion of linoleic acid (arrow) to give γ-linolenic acid (γ-18:3) by Ot-Des6.1.

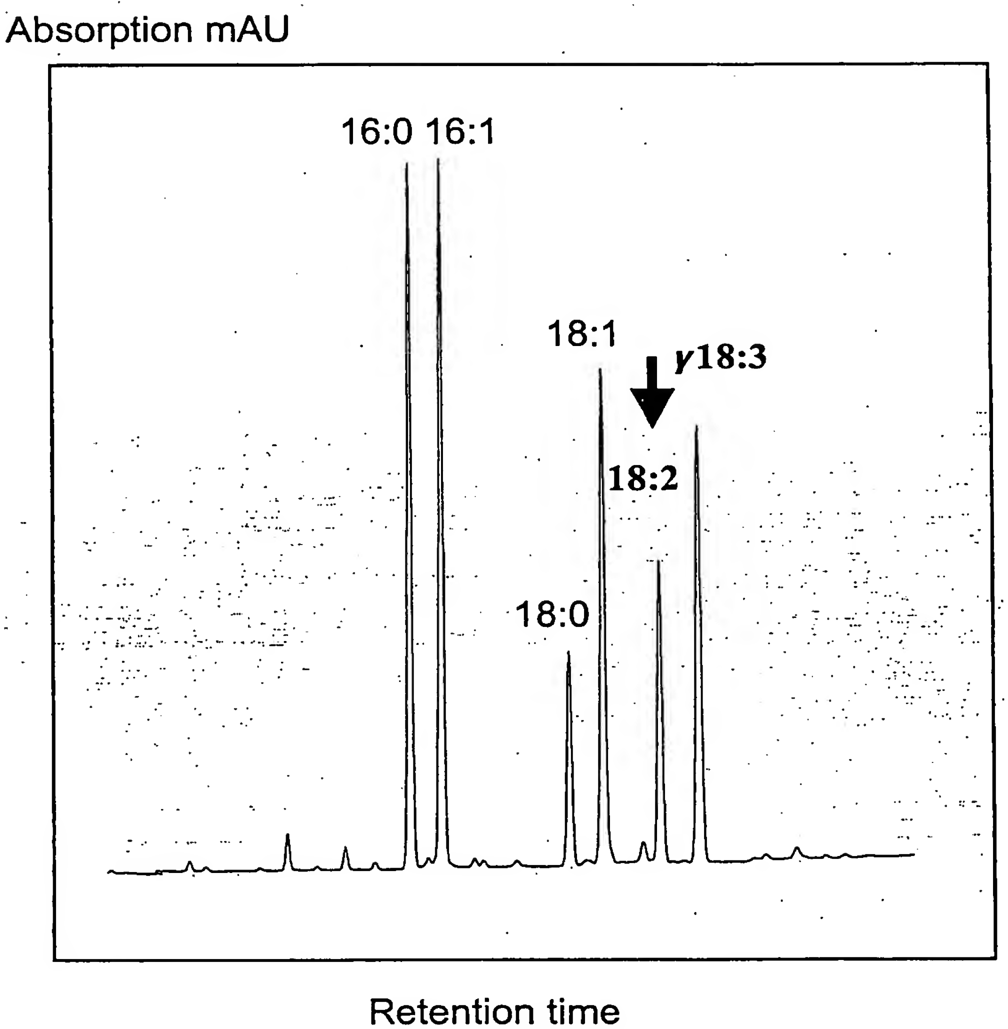
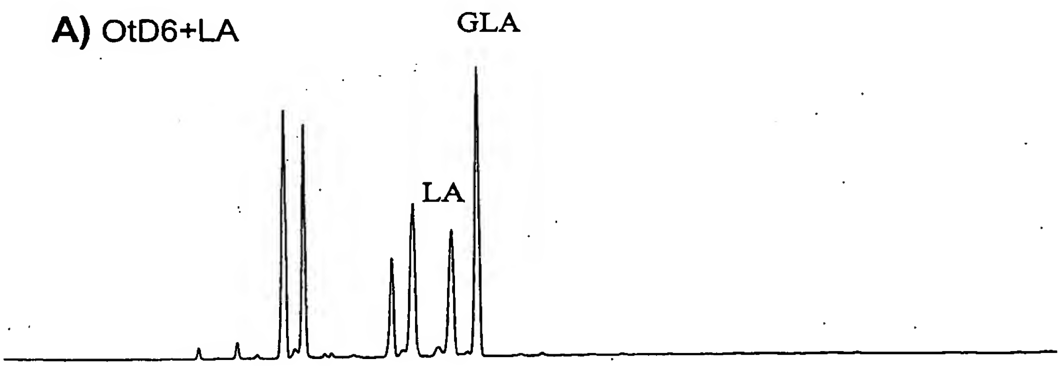
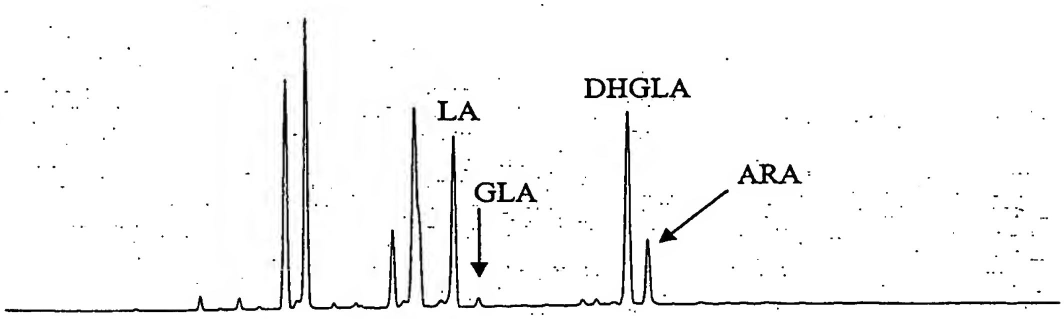


Figure 21: Conversion of linoleic acid and α-linolenic acid (A and C), and reconstitution of the ARA and EPA synthetic pathways, respectively, in yeast (B and D) in the presence of OtD6.1.

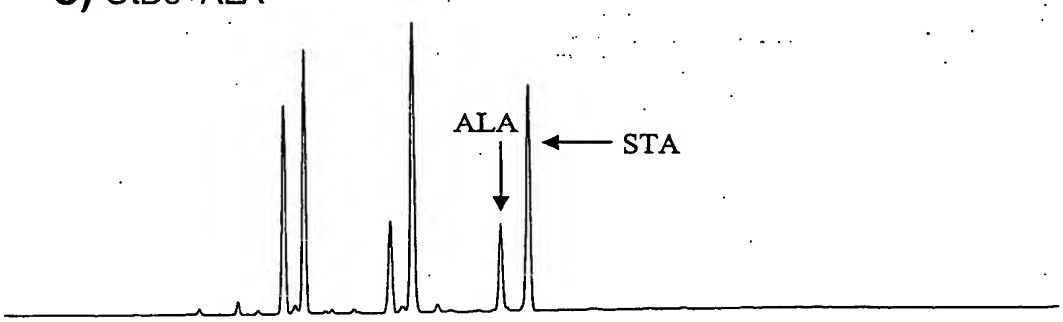
A) OtD6+LA



B) OtD6+PSE1+PtD5+LA



C) OtD6+ALA



D) OtD6+PSE1+PtD5+ALA

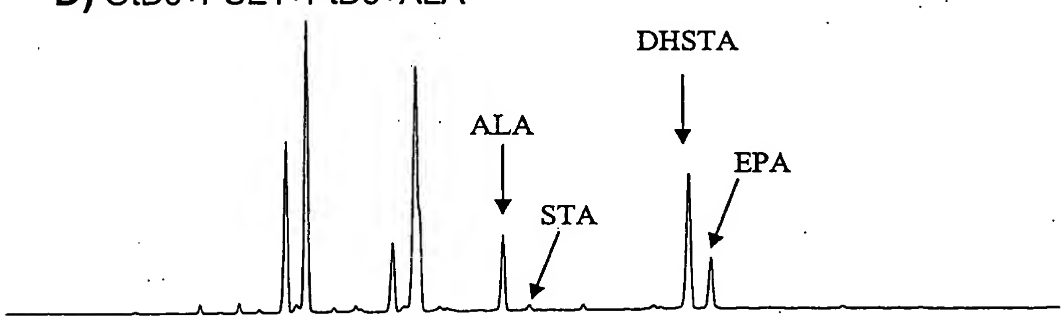


Figure 22: Expression of ELO(XI) in yeast

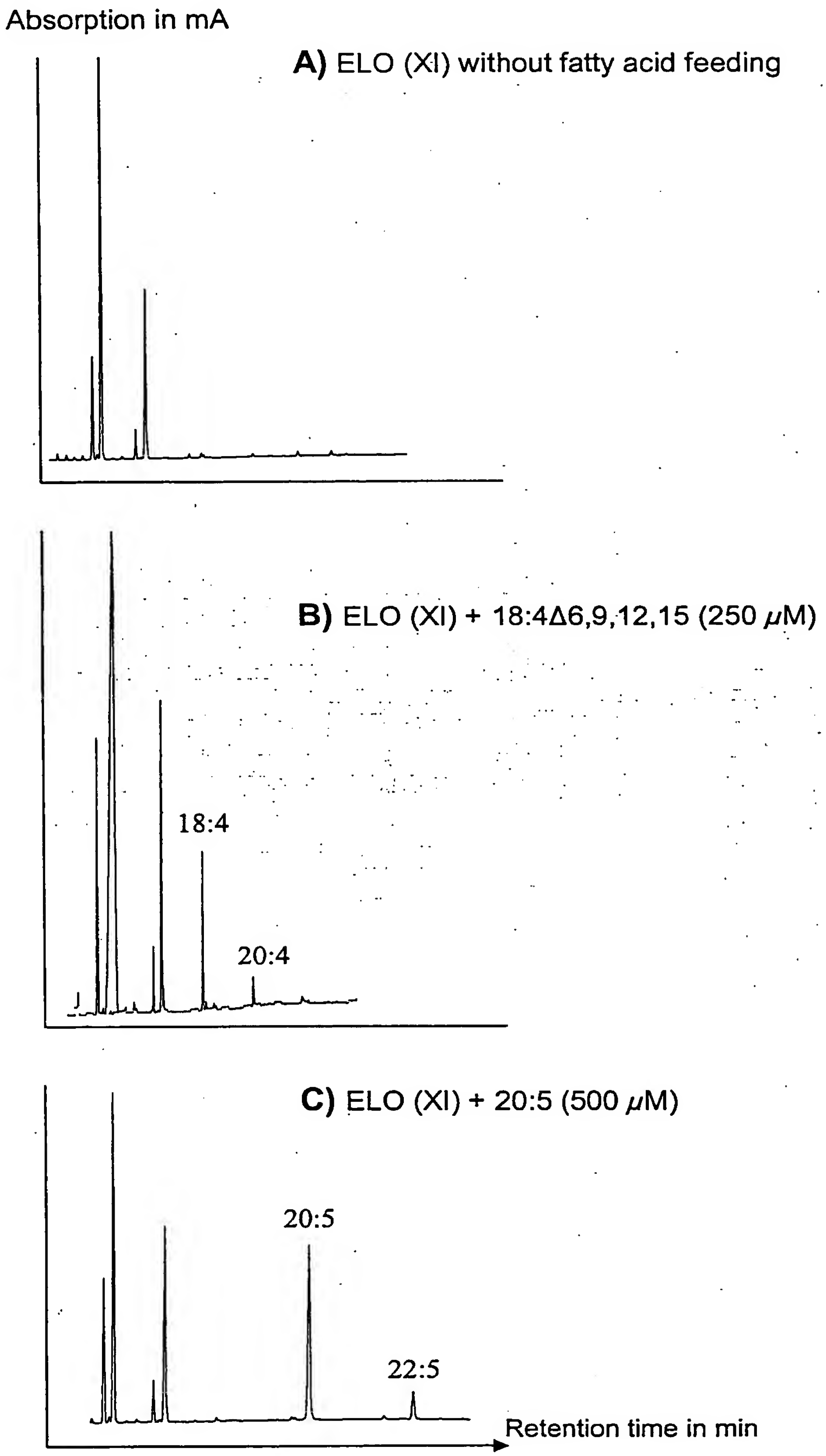


Figure 23:

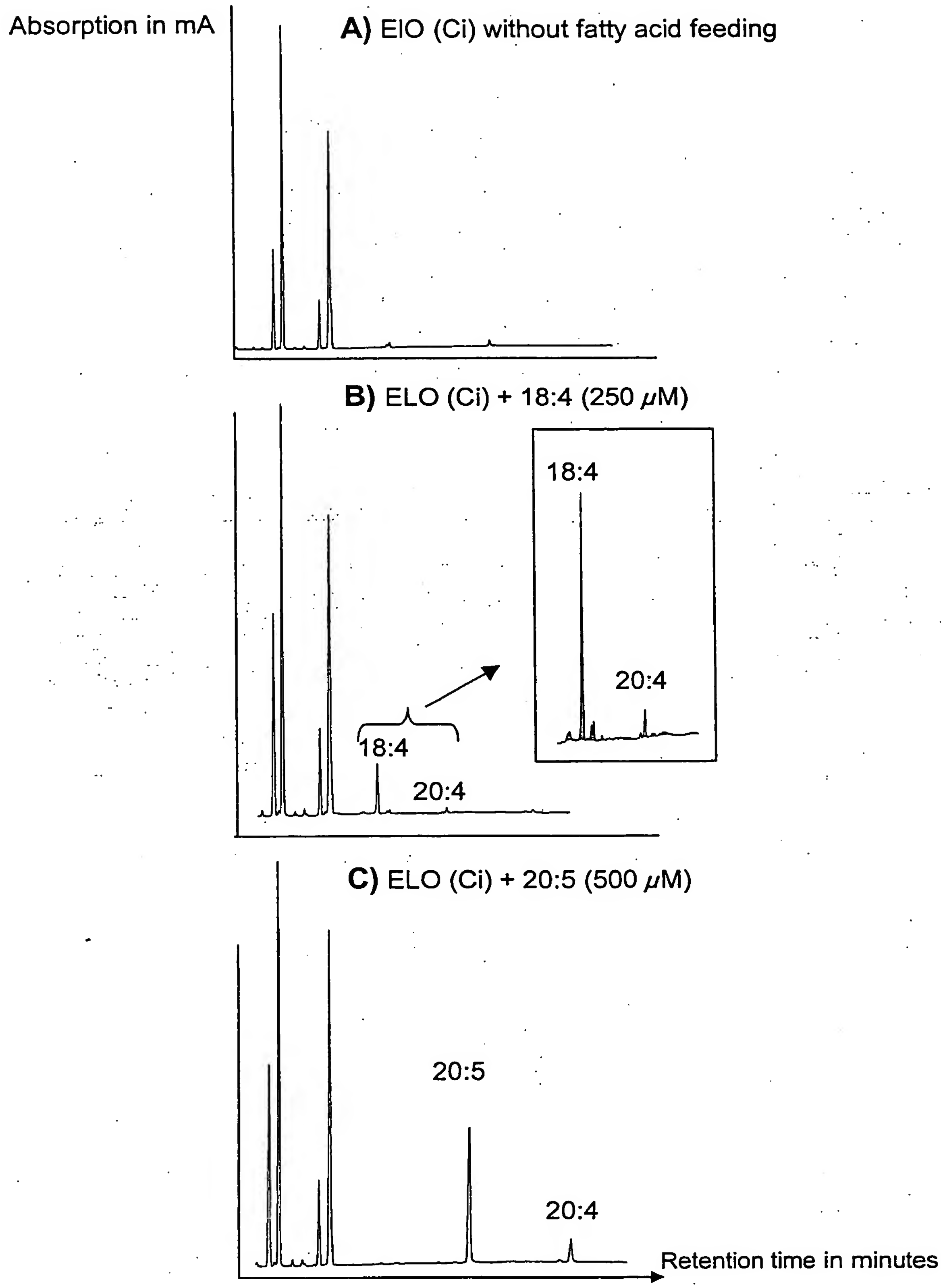

Absorption in mA
A) ElO (Ci) without fatty acid feeding
B) ELO (Ci) + 18:4 (250 μM)
18:4
20:4
18:4
20:4
C) ELO (Ci) + 20:5 (500 μM)
20:5
20:4
Retention time in minutes

Figure 24: Elongation of eicosapentaenoic acid by OtElo1 (B) and OtElo1.2 (D), respectively. The controls (A, C) do not show the elongation product (22:5ω3).

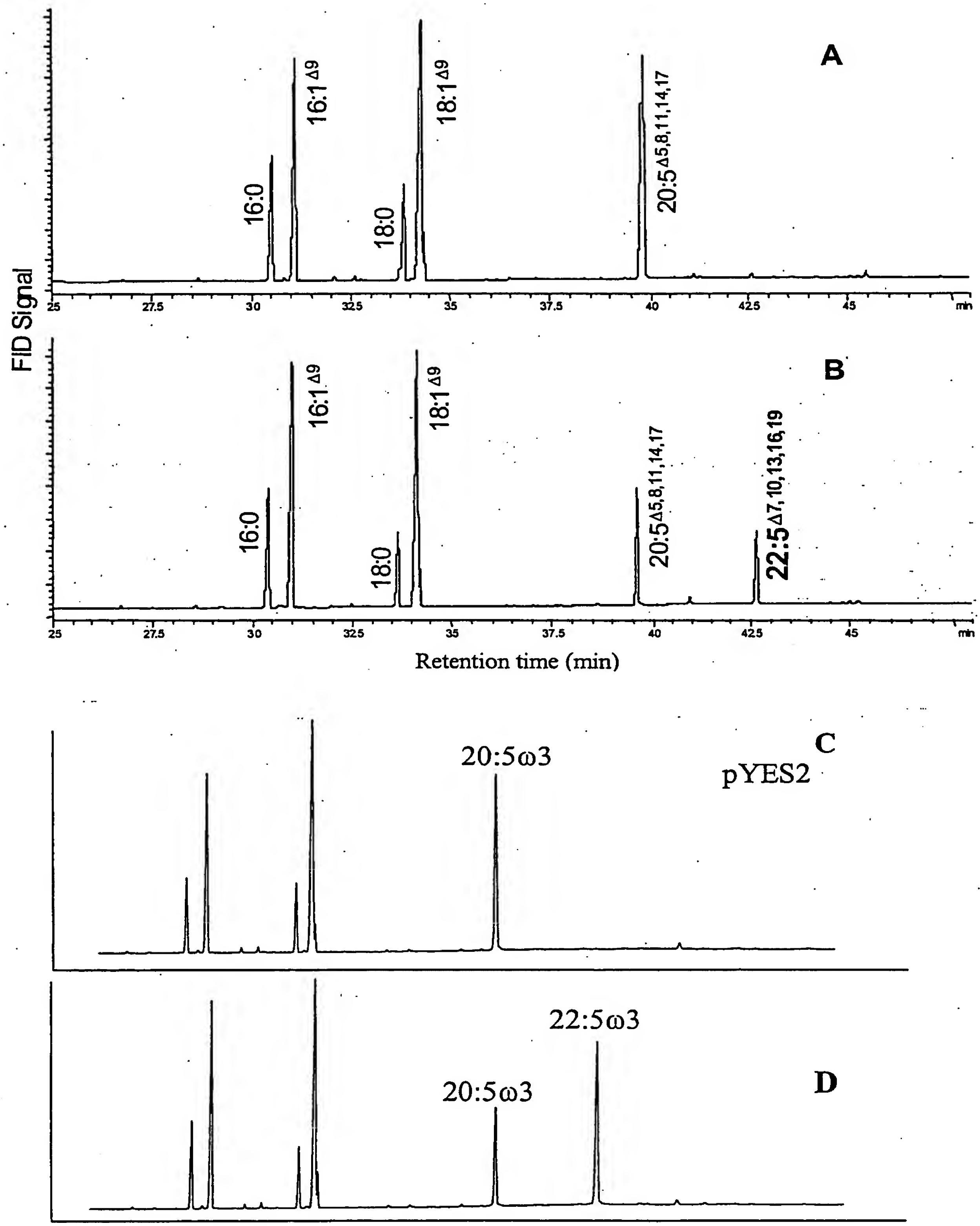


Figure 25: Elongation of arachidonic acid by OtElo1 (B) and OtElo1.2 (D), respectively. The controls (A, C) do not show the elongation product (22:4ω6).

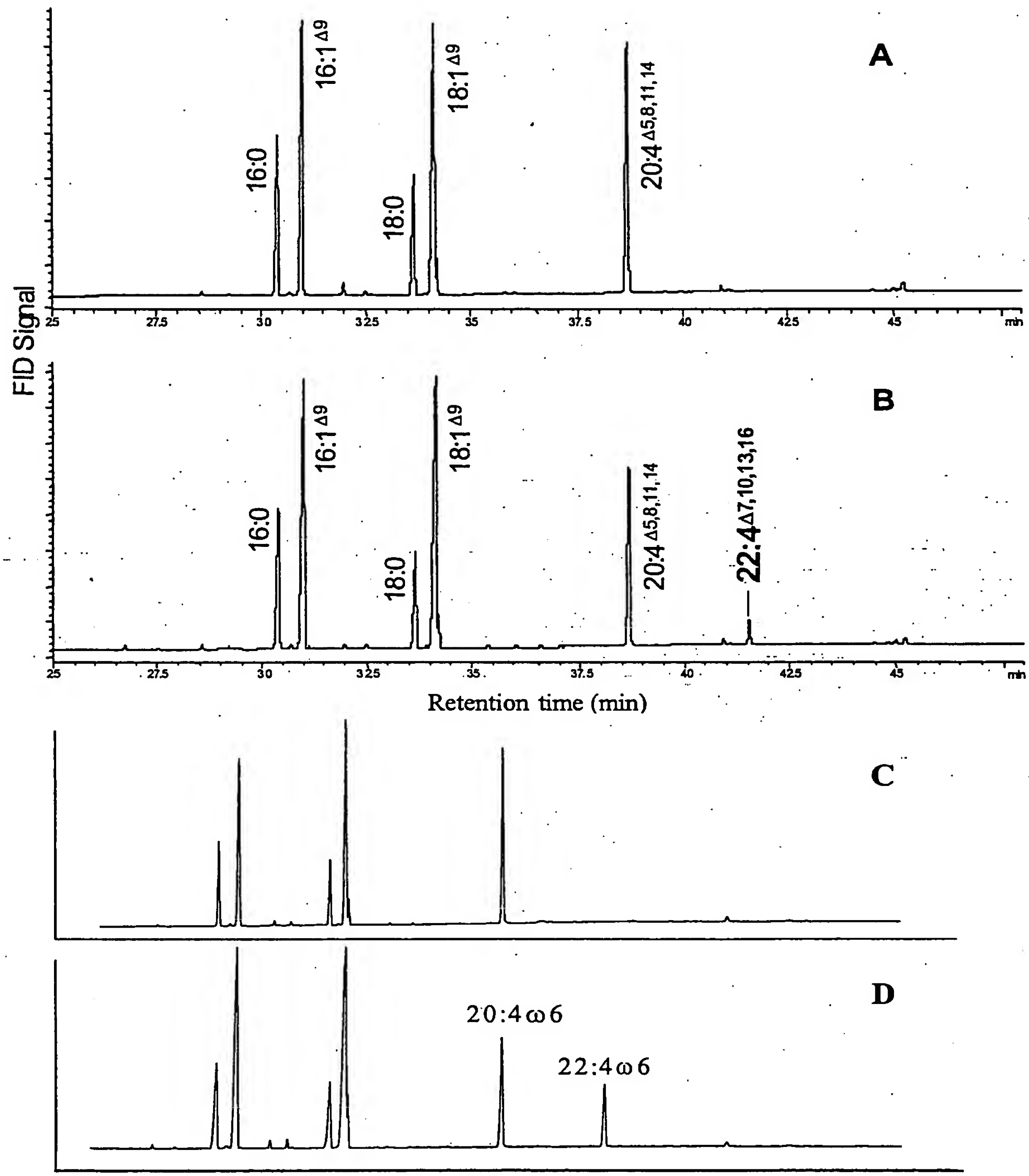


Figure 26: Elongation of 20:5n-3 by the elongases At3g06470.

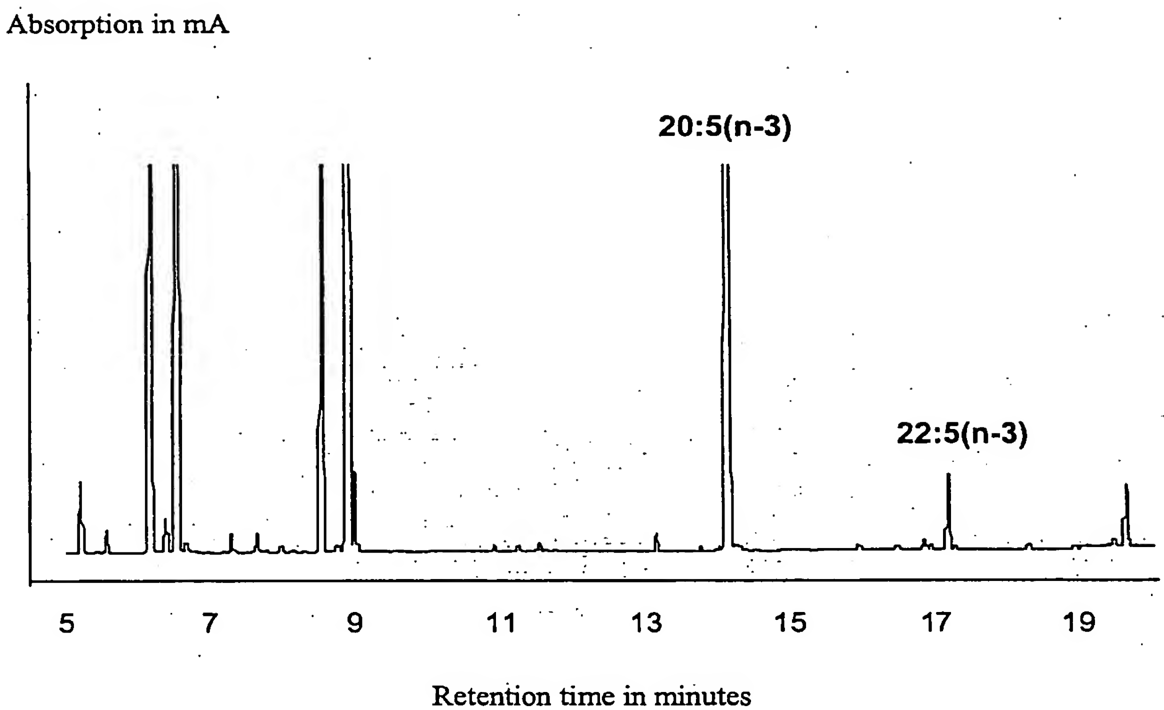


Figure 27: Substrate specificity of the Xenopus Elongase (A), Ciona Elongase (B) und Oncorhynchus Elongase (C)

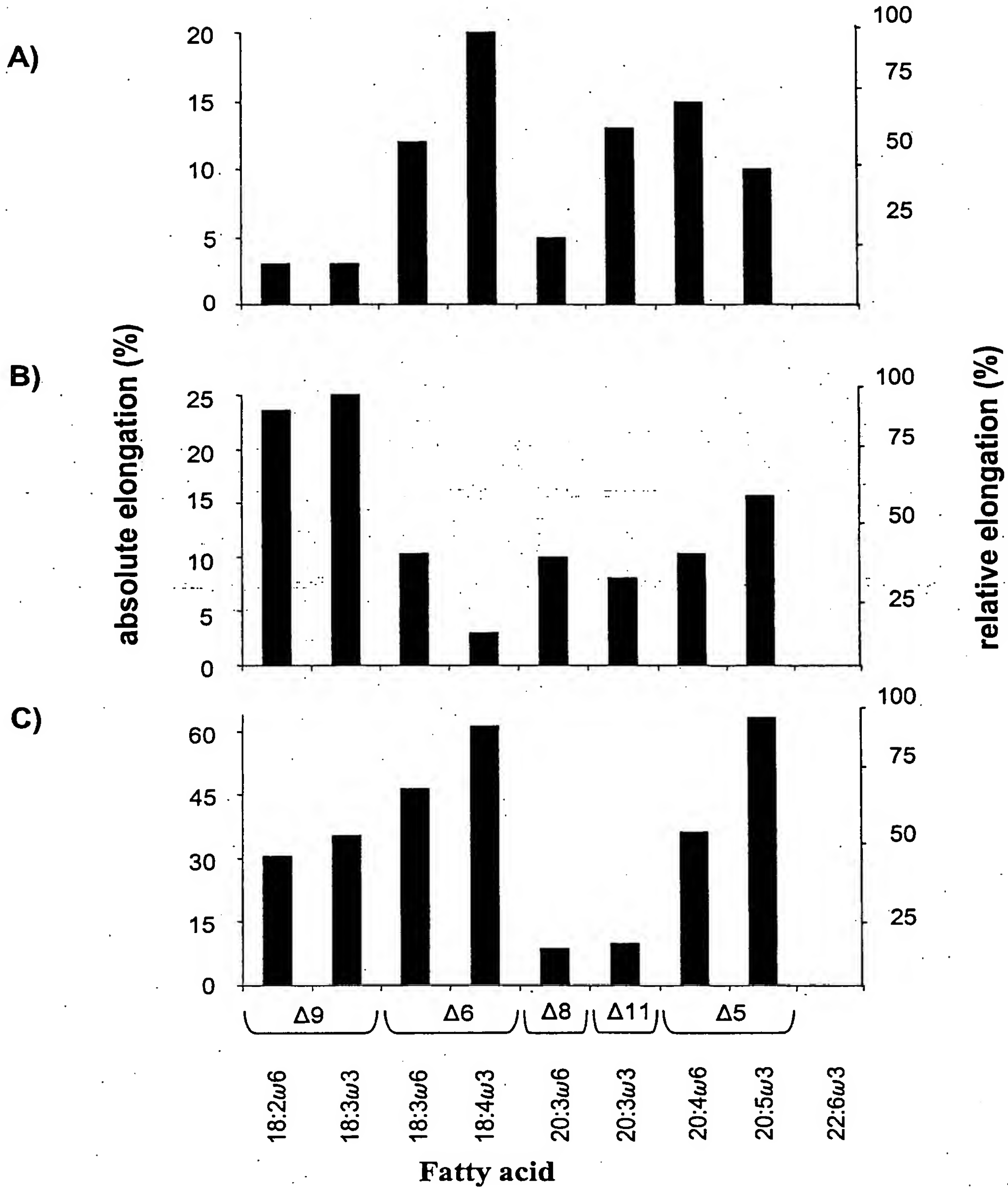


Figure 28: Substrate specificity of the Ostreococcus Δ5-elongase (A), the Ostreococcus Δ6-elongase (B), the Thalassiosira Δ5-elongase (C) and the Thalassiosira Ostreococcus Δ6-elongase (D)

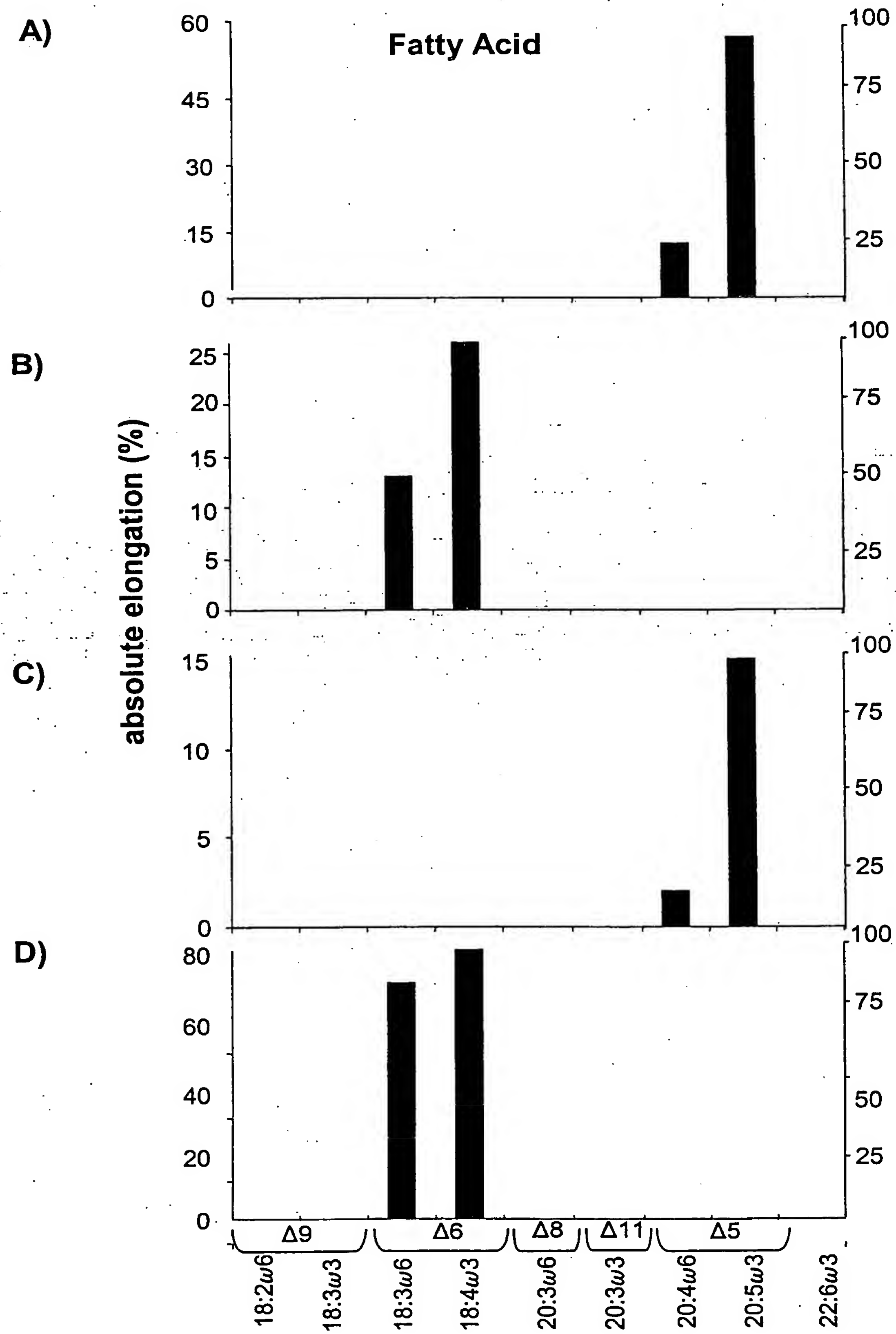
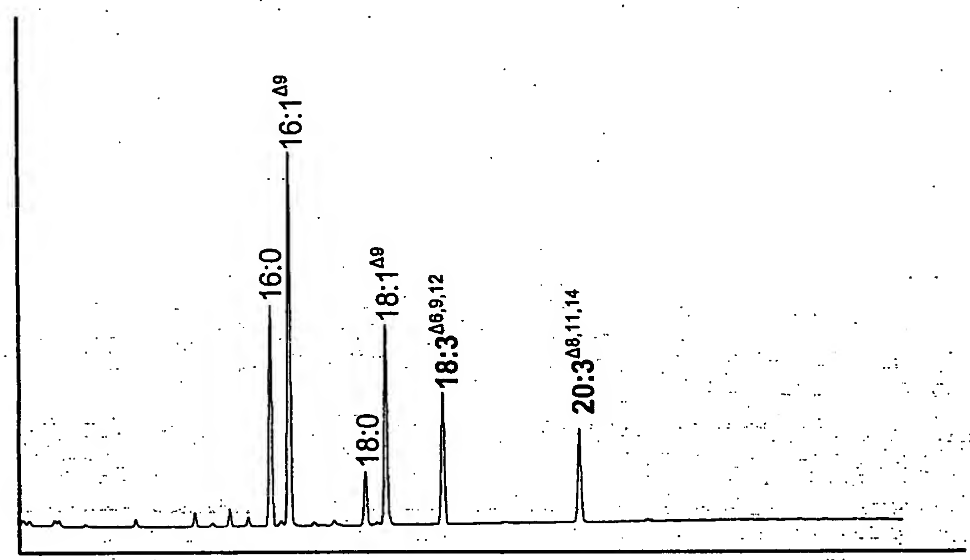


Figure 29: Expression of the Phaeodactylum tricornutum Δ6-elongase (PtELO6) in yeast. A) shows the elongation of the C18:3$^{\Delta 6,9,12}$ fatty acid and B) the elongation of the C18:3$^{\Delta 6,9,12,15}$ fatty acid

A)



B)

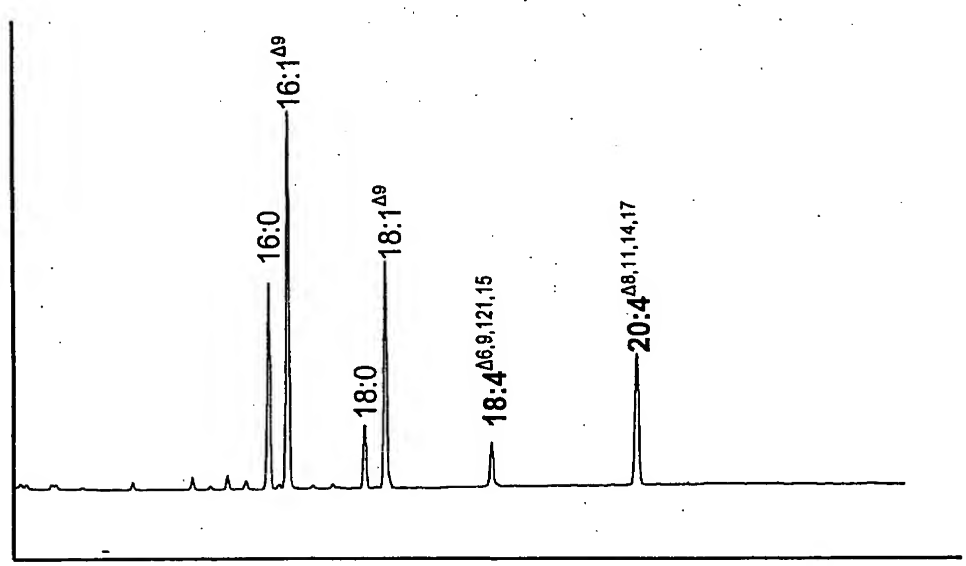


Figure 30: Figure 30 shows the substrate specificity of PtELO6 with regard to the substrates fed.

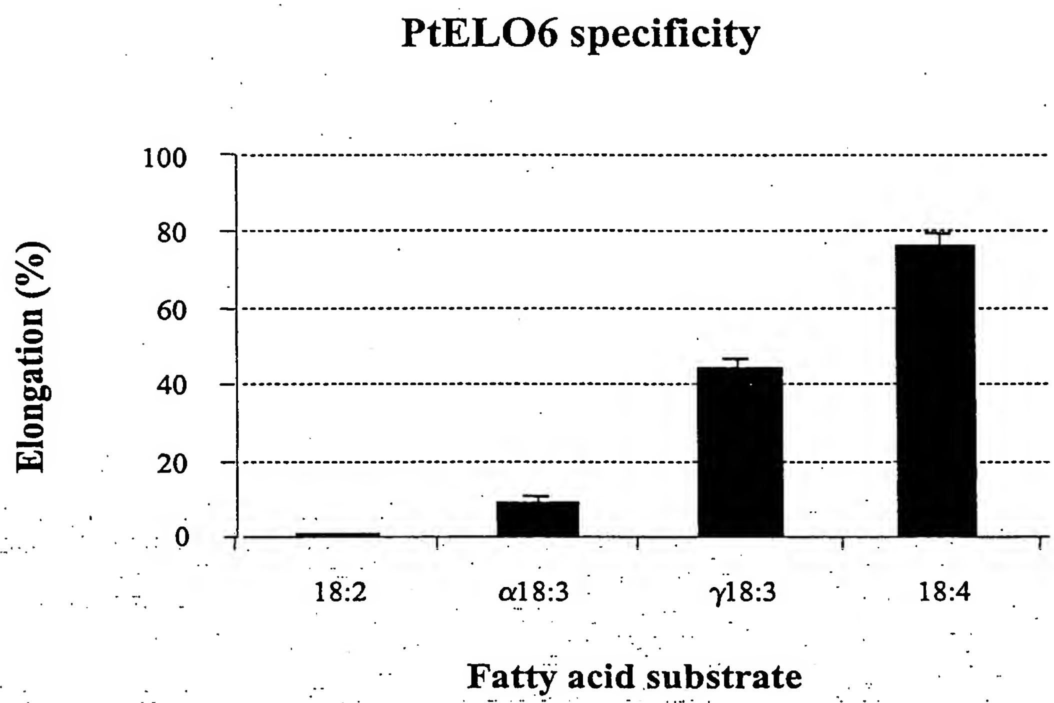


Figure 31: Gas-chromatographic analysis of the seed of a transgenic plant, transformed with pSUN-5G.

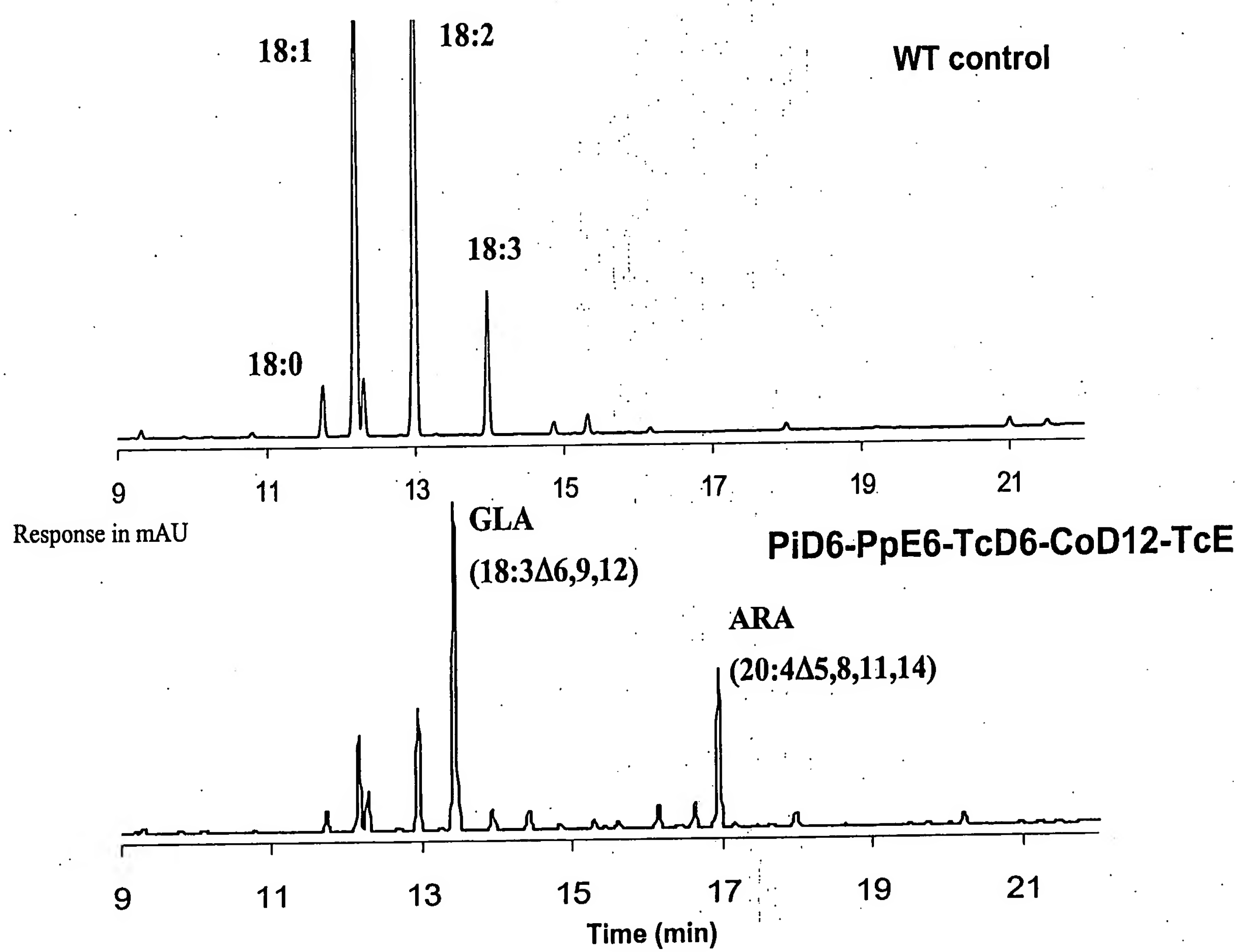


Figure 32: Gas-chromatographic analysis of the seed of a transgenic plant, transformed with pGPTV-D6Des(Pir)_D5Des(Tc)_D6Elo(PP)_12Des(Co)

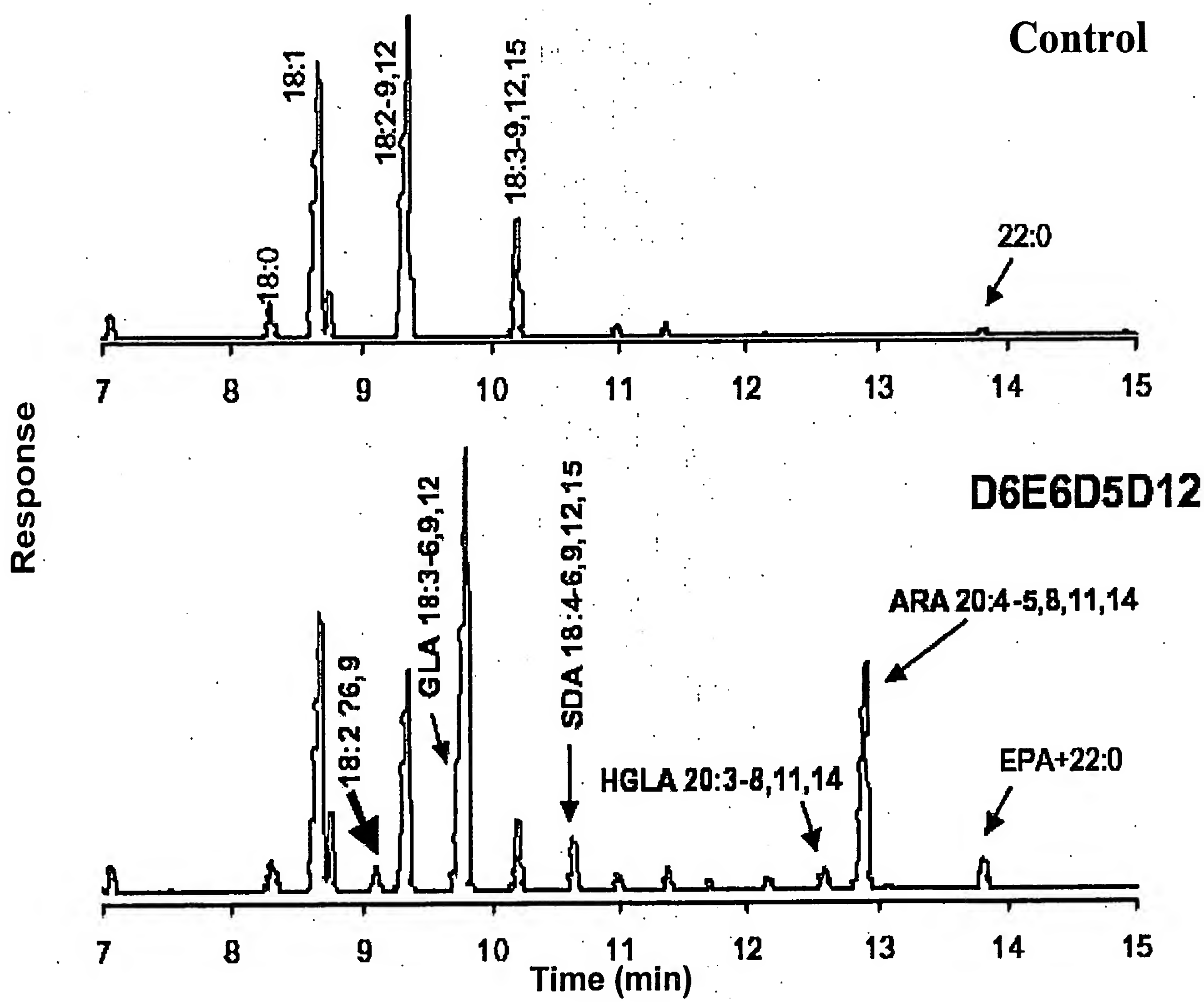


Figure 33: DHA in transgenic seeds of Brassica juncea. The plants were transformed with the construct pSUN-8G.